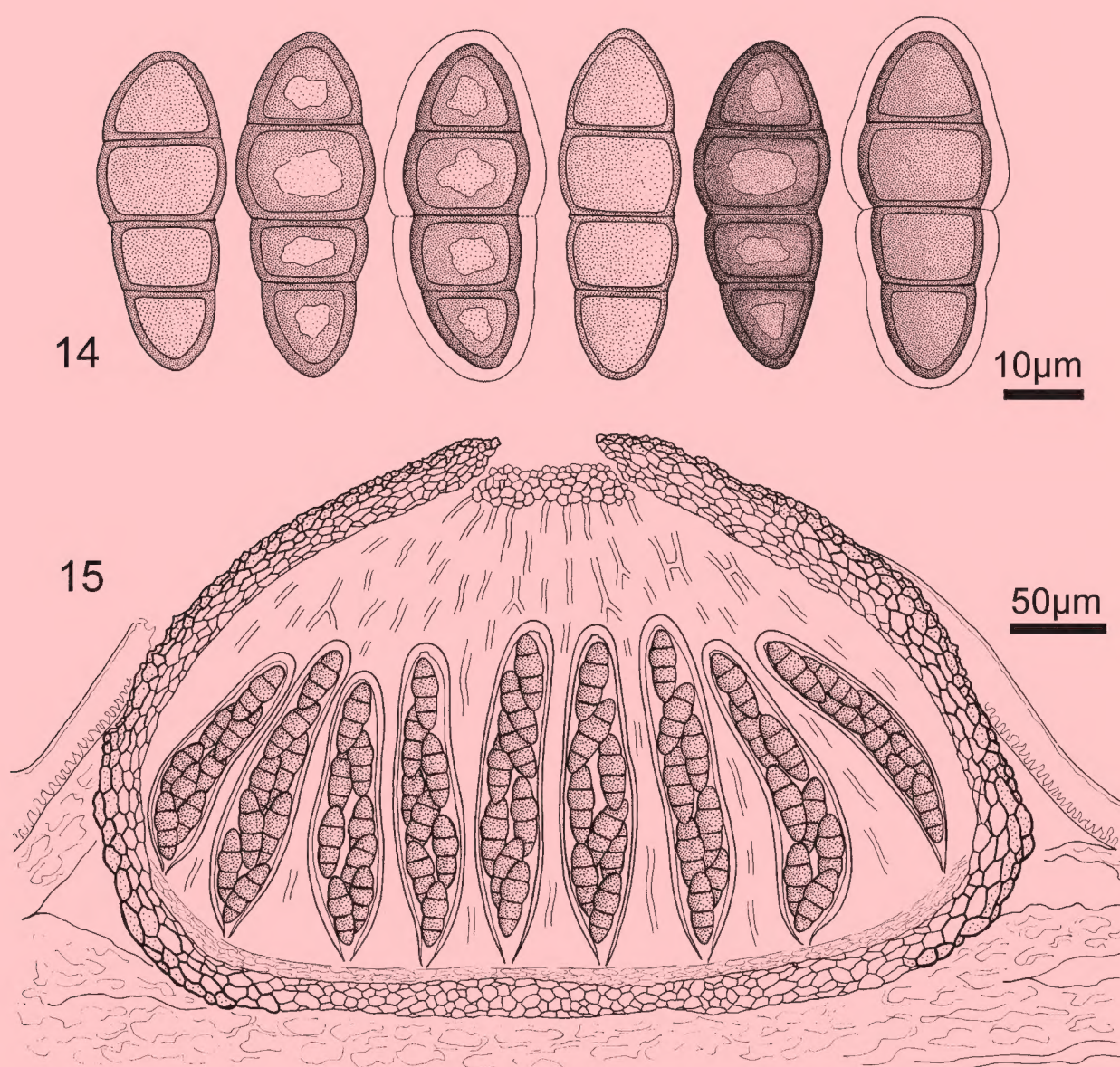


# MYCOTAXON

THE INTERNATIONAL JOURNAL OF FUNGAL TAXONOMY & NOMENCLATURE

VOLUME 113

JULY–SEPTEMBER 2010



Tanaka, Hirayama & Iqbal  
FIGS 14–15. *Diadema ahmadii* sp. nov.  
(p. 340)





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THE INTERNATIONAL JOURNAL OF FUNGAL TAXONOMY & NOMENCLATURE

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## MYCOTAXON

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**Genera of *Pezizales* of Argentina 1.  
An updating of selected genera**

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**Abstract** — Twenty-two genera of *Pezizales* from Argentina belonging to families *Discinaceae*, *Helvellaceae*, *Morchellaceae*, *Pezizaceae*, *Pyronemataceae*, *Sarcoscyphaceae*, and *Sarcosomataceae* are reviewed according to new nomenclatural and taxonomical parameters. Some changes in type species selection are noted and relationships among genera based on microscopical, ultramicroscopical, and molecular data are discussed. The related anamorphs, when known, are briefly described.

**Resumen** — Se revisan y actualizan veintidós géneros de *Pezizales* de Argentina pertenecientes a las familias *Discinaceae*, *Helvellaceae*, *Morchellaceae*, *Pezizaceae*, *Pyronemataceae*, *Sarcoscyphaceae* y *Sarcosomataceae* de acuerdo con nuevos conceptos taxonómicos y nomenclaturales. Se incluyen algunos cambios en la designación de las especies tipos con respecto a trabajos anteriores de la autora y comentarios de las relaciones filogenéticas entre los géneros, considerando datos microscópicos, ultramicroscópicos y moleculares. Se describen brevemente los anamorfos de cada género, cuando se los conoce.

**Key words** — *Ascomycota*, cup-fungi, taxonomy, biodiversity

**Introduction**

About fifty years after the my first publication on discomycetes of Argentina, I thought that perhaps it would be worthwhile to produce an update of my work on the taxonomy of this group, mainly regarding current concepts and nomenclature of the genera.

During this time, generic concepts have been enriched and refined by the use of modern tools such as scanning electron microscopy (SEM) and transmission electron microscopy (TEM). Moreover, molecular studies have helped clarify relationships between taxa, leading to hypothetical phylogenies.

Advances in the nomenclature of various discomycete taxa have also required changes to some generic and specific names used in my previous papers. However, as generic limits differ from author to author, I am giving my views and provide generic descriptions that cover my concepts.

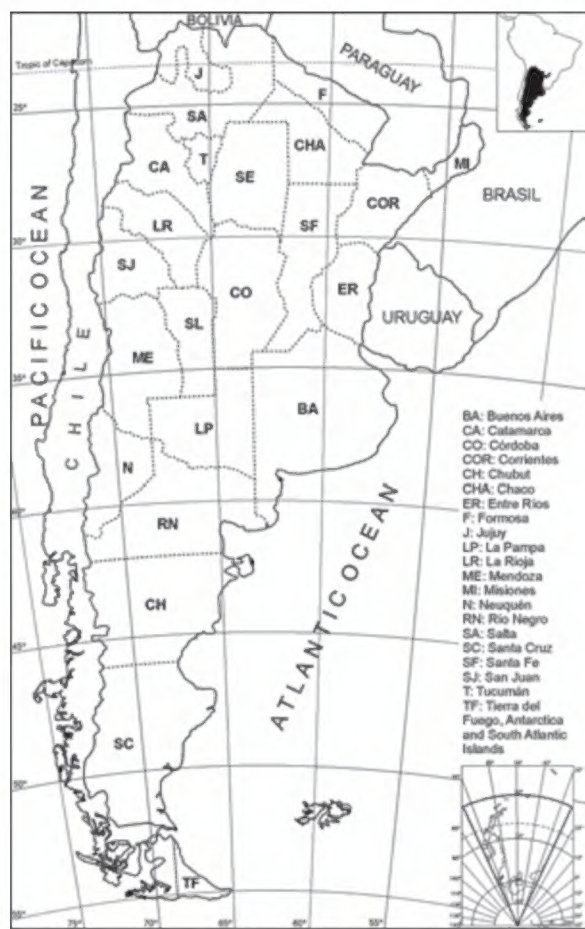


FIG. 1. Map of Argentina

Argentina is an extensive country covering an area of 2,766,891 km<sup>2</sup>, extending from 21°46'S to 55°03'S and 53°38'W to 73°35'W. The altitude decreases from W to E, ranging from the Andes, whose highest point is 6,962 m to sea level at the Atlantic coast, but with its lowest depression (−105m) in the Patagonian plateau. It encompasses different climates, from subtropical in the north, temperate in the center and south, and to polar if the Argentine Sector of Antarctica is included. The mycobiota is therefore very diverse. So far, many territories are still poorly explored regarding the discomycete biota and many genera and species are yet to be discovered.

At present, there are approximately 37 genera of *Pezizales* recognized from Argentina. I refer only to 22 in this contribution, choosing those that have been recently monographed or studied with ultrastructural or molecular tools. The remaining shall be treated in a next contribution.

The genera are presented in alphabetical order and include a description, type species, habitat, geographical distribution in Argentina, notes on related genera, and a bibliography. Abbreviated literature is presented at the end of the generic descriptions and full references can be found in the literature. Each genus is illustrated with a species recorded in Argentina, which is depicted in a plate (PLATES 1–22). A map (FIG. 1) shows the provinces with the abbreviations listed in the accompanying legend. Taxonomic categories above genera follow Kirk et al. (2008) and the website “Index Fungorum” (<http://www.indexfungorum.org/BSM./bsm.asp>). Electronic libraries such as Cybertruffle (<http://cybertruffle.org>).



[uk/cyberliber](http://www.cyberliber.com.ar)) and Biblioteca electrónica del Ministerio de Ciencia y Tecnología de la Argentina (<http://www.biblioteca.mincyt.gov.ar>) were very valuable information sources for this paper.

Ultrastructural references include Bellemère et al. (1990), Kimbrough (1994), Kimbrough & Curry (1986), Kimbrough & Gibson (1989, 1991), Kimbrough et al. (1990), Li & Kimbrough (1995, 1996a,b) and Meléndez-Howell et al. (2003). Molecular data were extracted from Hansen et al. (1999, 2001), Hansen & Pfister (2006), Harrington et al. (1999), Landvik et al. (1997, 1999), Læssøe & Hansen (2007), Liu & Zhuang (2006), O'Donnell et al. (1997), Perry et al. (2007), Perry & Pfister (2008), Tedersoo et al. (2006), and Weinstein et al. (2002).

## Taxonomy

*Acervus* Kanouse emend. Pfister (*Pyronemataceae*)

ASCOMATA apothecial, medium-sized, superficial, sessile to subsessile, at first globose becoming shallow cupulate (cleistohymenial), scattered to gregarious, sometimes concrescent forming masses of several cm across arising from a mycelium agglomerated in a dense mat mixed with the substratum, firm fleshy consistency; disc bright yellow to orange, the pigment soluble in water and alcohol; margin entire or lobate, reflexed and undulate; external surface concolorous with the disc or paler, pruinose to furfuraceous. ECTAL EXCIPULUM of textura globulosa to angularis composed of isodiametric cells, the most superficial smaller than the internal ones and containing orange granules, bearing flexuous, cylindrical, short, obtuse hair with few septa. MEDULLARY EXCIPULUM well developed, of a lax textura intricata composed of hyaline hyphae with swollen articles. SUBHYMENIUM of dense textura globulosa, the cells containing pigment and smaller than those of the excipulum. ASCI cylindrical, 8-spored, J–, dehiscence indistinct. PARAPHYSES robust, cylindrical, subclavate or irregularly enlarged, containing pigmented granules near the apex, pluriseptate. ASCOSPORES uninucleate, 1-seriate, multiguttulate, hyaline, broad ellipsoidal with blunt ends to subglobose, smooth, thin-walled.

TYPE SPECIES: *Acervus aurantiacus* Kanouse Pap. Mich. Acad. Sci. 23:149. 1938 [= *A. epispartius* (Berk. & Broome) Pfister].

HABITAT: on damp soil, sometimes among grass, rotten wood and debris.

ANAMORPH: unknown.

NOTES: *Acervus* is an earlier synonym of *Phaedropezia* Le Gal. It shares with *Caloscypha* Boud. a bright orange-yellow disc and the same type of septal structure. TEM studies of the hyphae revealed that a membrane-like translucent band borders the pore plug where the Woronin bodies are crystalloid, a view that supports the inclusion of both genera in the same tribe of the *Pyronemataceae*. However, *Caloscypha* differs in the ascomata that turn green or bluish with age or when touched or broken. *Ascosparrassis* Kobayasi is similar to *Acervus*



in its small, guttulate ascospores, robust paraphysis, and orange ascoma of sparassoid habit. Formerly *Acervus* was placed in the *Sarcosomataceae* or *Sarcoscyphaceae* because asci were considered suboperculate. Other authors held the view that dehiscence is somewhat bilabiate, as in *Caccobius* Kimbr. and *Thelebolus* Tode (*Thelebolaceae*). Nevertheless operculate asci can be observed in the Argentine collection of *A. epispartius* (PLATE 1, FIG. 3). The fact that ascospores are uninucleate instead of multinucleate reinforces the view that *Acervus* belongs to the *Pyronemataceae*. This family is considered here in a wider sense than in Kimbrough's (1989) proposal (in accordance with Kirk et al. 2008). Molecular phylogenetic studies show discrepancies in the concept of *Pyronemataceae*. Some authors consider it monophyletic, others think it is polyphyletic, but they agree that *Acervus* occupies an isolated place that forms a separate monophyletic group.

DISTRIBUTION IN ARGENTINA: only one collection of *A. epispartius* — cited as *A. aurantiacus* — has been found in Argentina from BA.

ILLUSTRATION: Pl. 1, 1–6. *Acervus epispartius*.

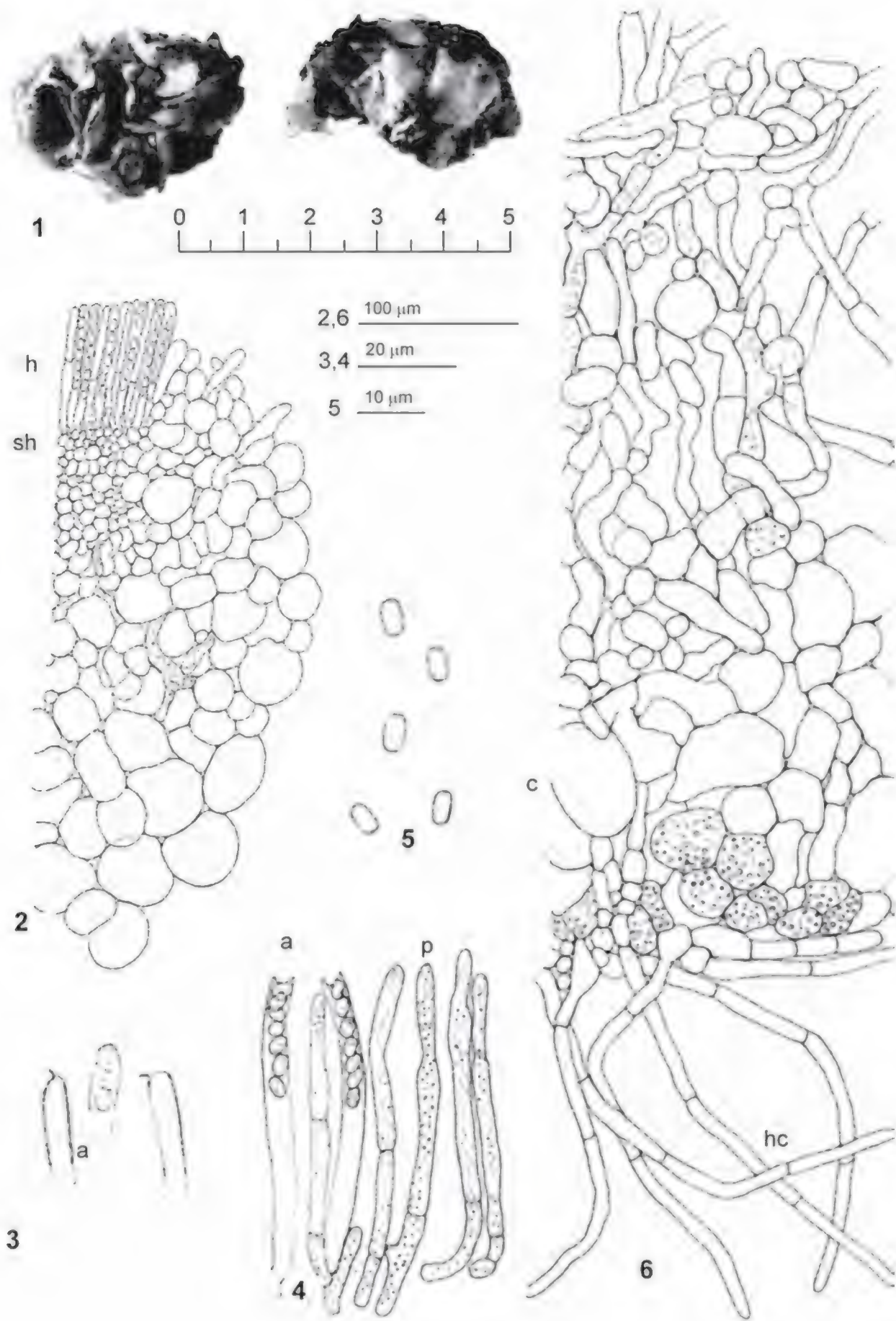
LITERATURE: Eckblad 1968; Gamundí 1970; Kimbrough 1989; Kimbrough & Curry 1986; Kirk et al. 2008; Korf 1963, 1988; Le Gal 1953; Liu & Zhuang 2006; Moravec 1983; Perry et al. 2007; Pfister 1975; Pfister & Bessette 1985; Pfister & Halling 1989; Zhuang & Wang 1998.

### *Aleuria* (Pers.) Fuckel (*Pyronemataceae*)

ASCOMATA apothecial, small to large, up to 8 cm diam., superficial, sessile to subsessile, cupuliform to cochleate, scattered, gregarious or cespitose, bright coloured; disc smooth, yellow, orange to reddish orange, margin conspicuous; external surface paler than the disc, whitish in dried specimens, pruinose, furfuraceous to tomentose. ECTAL EXCIPULUM of textura angularis to textura globulosa of isodiametric or elongated cells, the external ones ending in superficial, hyphoid, short, obtuse, hyaline hairs. MEDULLARY EXCIPULUM of textura intricata composed of hyaline hyphae densely arranged. Septal pores of excipular cells (TEM) have a lamellate structure and globose Woronin bodies are associated with them. SUBHYMENIUM an orange-yellow zone of textura angularis, of small cells. ASCI cylindrical, 8-spored, J–. PARAPHYSES pluriseptate, subclavate or bent at the apex, containing granules of carotenoids (major pigments  $\beta$ - and  $\gamma$ -carotene, ester of aleuriaxanthine) that turn green with iodine. ASCOSPORES uninucleate, 1-seriate, containing 1–2 guttules, hyaline to pale yellowish, ellipsoidal, with a conspicuous cyanophilic ornamentation ridge- or net-like, sometimes forming apicula at both ends or with prominent pointed warts.

---

PLATE 1. 1–6. *Acervus epispartius* (LPS 35273). 1. Concretescent ascomata: frontal and lateral view. 2. Vertical section of the ascoma: h, hymenium, sh subhymenium. 3. Ascus apex. 4. Hymenium: a, mature asci, p, paraphyses. 5. Ascospores. 6. Vertical section at the base of the ascoma: c, excipulum, hc, basal hyphae.



TYPE SPECIES: *Aleuria aurantia* (Pers.) Fuckel, Jahrb. Nassauischen Vereins. Naturk. 23–24: 325. 1870.

HABITAT: on sandy soil, rich forest soil or gravy soil along paths, frequently on disturbed sites, sometimes among grass or mosses.

ANAMORPH: unknown.

NOTES: The cosmopolitan species *Aleuria aurantia* is commonly named orange peel peziza. The genus is close to *Melastiza* due to its mostly reticulate ascospores and paraphyses containing the same carotenoid pigments but differs in the external surface of the apothecium. This similarity led Moravec to unite them under the older name, *Aleuria*, with two subgenera *Aleuria* and *Melastiza*. At least one of Moravec's arguments (i.e., 'the same habitat'; see NOTES under *Melastiza*) for merging both genera is dubious. *Rhodopeziza* is also similar, sharing the coloured hymenium and the cyanophilic ascospore ornamentation, but differs in the weak J+ ascus wall reaction (see NOTES under *Rhodopeziza*). TEM studies of septal structure in the ascus cell and the ascogenous hyphae show a granular opaque matrix, which borders the pore, appearing in older asci as a fan-shaped plug with a lamellate electron-translucent torus adjacent to the pore rim (referred to as the 'aleurioid' type). A phylogram derived from SSU rDNA sequences suggests that *Aleuria* is related to *Byssonectria* P. Karst. and forms a group containing the genera *Scutellinia*, *Cheilymenia*, and *Pyronema* Carus (Landvik et al. 1997). This clade is not supported by another study based on nLSU rDNA sequences (Perry et al. 2007). It appears that the presence of carotenoid pigment is of little phylogenetic significance.

DISTRIBUTION IN ARGENTINA: *A. aurantia* is the only species recorded, distributed in BA, N, RN, SC, TF.

ILLUSTRATION: Pl. 2, 1–6. *Aleuria aurantia*.

LITERATURE: Arpin 1969; Gamundí 1960, 1975; Gamundí & Horak 2003; Gamundí et al. 2004; Häffner 1993; Kaushal 1976; Kimbrough 1989, 1994; Kimbrough & Curry 1986; Landvik et al. 1997; Liu & Zhuang 2006; Moravec 1972, 1994a; Perry et al. 2007; Rifai 1968; Spooner & Yao 1995.

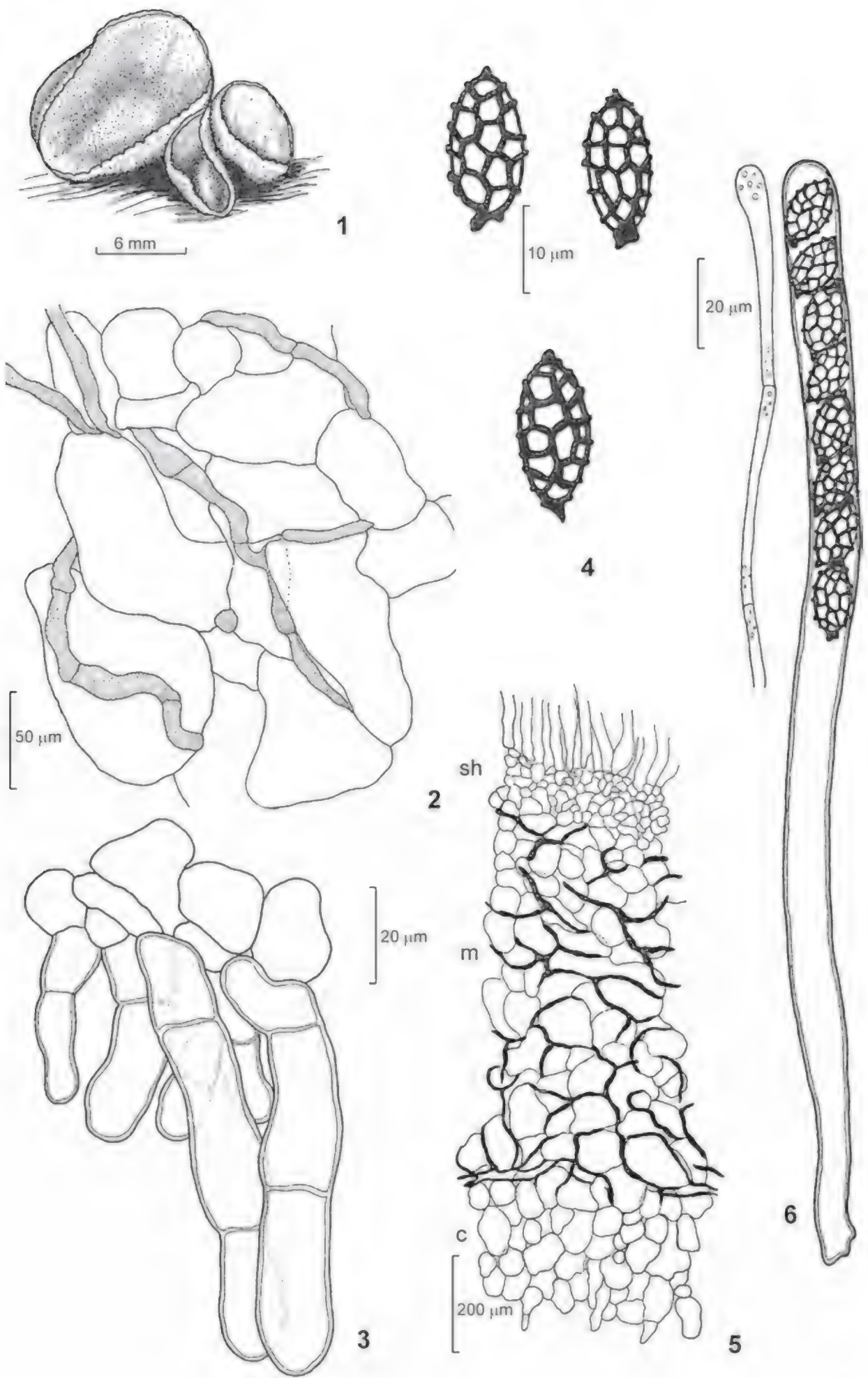
### *Aleurina* Massee (*Pyronemataceae*)

ASCOMATA apothecial, medium-sized, superficial, sessile to subsessile, scattered to gregarious, cup shaped to discoid at maturity, the base with abundant subhyaline hyphae often enmeshing soil particles; disc olivaceous, brown to

---

PLATE 2. 1–6. *Aleuria aurantia* (BAFC 21059). Ascomata. 2. Medullary excipulum. 3. Receptacle: surface hairs. 4. Ascospores. 5. Vertical section of the ascoma: sh, subhymenium, m, medullary excipulum, c, ectal excipulum. 6. Ascus and paraphyses.





purplish brown; external surface brown to reddish brown, smooth but pustulate near the margin. ECTAL EXCIPULUM a textura angularis of isodiametric to elongated polygonal light brown cells disposed at right angle to the surface, the most superficial smaller, subglobose with thick dark brown walls, aggregated to form the marginal pustules, sometimes with an extra inner layer of small cells with dark brown walls. MEDULLARY EXCIPULUM of textura intricata, composed of pale brown hyphae running horizontally. SUBHYMENIUM of compact textura intricata. ASCI cylindrical, 4- or 8-spored, J-. PARAPHYSES subcylindrical, subclavate or subcapitate, containing a dark, opaque, brown pigment at the apex, septate. ASCOSPORES uninucleate, 1-seriate, mostly 2-guttulate, hyaline to pale yellow, ellipsoidal, ornamented with cyanophlic conical or rounded warts or spines.

TYPE SPECIES: *Aleurina tasmanica* Masee, Bull. Misc. Inf., Kew 1898.

HABITAT: on soil sometimes among bryophytes, wood, or duff.

ANAMORPH: unknown.

NOTES: *Aleurina* is an earlier synonym of *Jafneadelphus* Rifai (1968). It is close to *Jafnea* Korf emend. Rifai in the structure of the ectal excipulum but this genus has superficial brown hairs, a cushion-like pseudostipe, and fusoidal to fusiform-ellipsoidal ascospores. It is also distinct from *Eoaleurina* Korf & W.Y. Zhuang characterized by the ectal excipulum of textura globulosa to angularis with cells of thin, hyaline walls, the most superficial with pigmented cytoplasm. *Smardaea* Svrček differs in the presence of a purplish, water-soluble pigment in the medullary excipulum. A phylogenetic analysis based on LSU rDNA sequences places *Aleurina* in a group that includes *Smardaea*.

DISTRIBUTION IN ARGENTINA: Two species are recorded: *A. argentina* (Rifai) Korf & W.Y. Zhuang, and *A. echinata* (Gamundí) Korf & W.Y. Zhang from N, RN, TF.

ILLUSTRATION: Pl. 3, 1–8. *Aleurina echinata*.

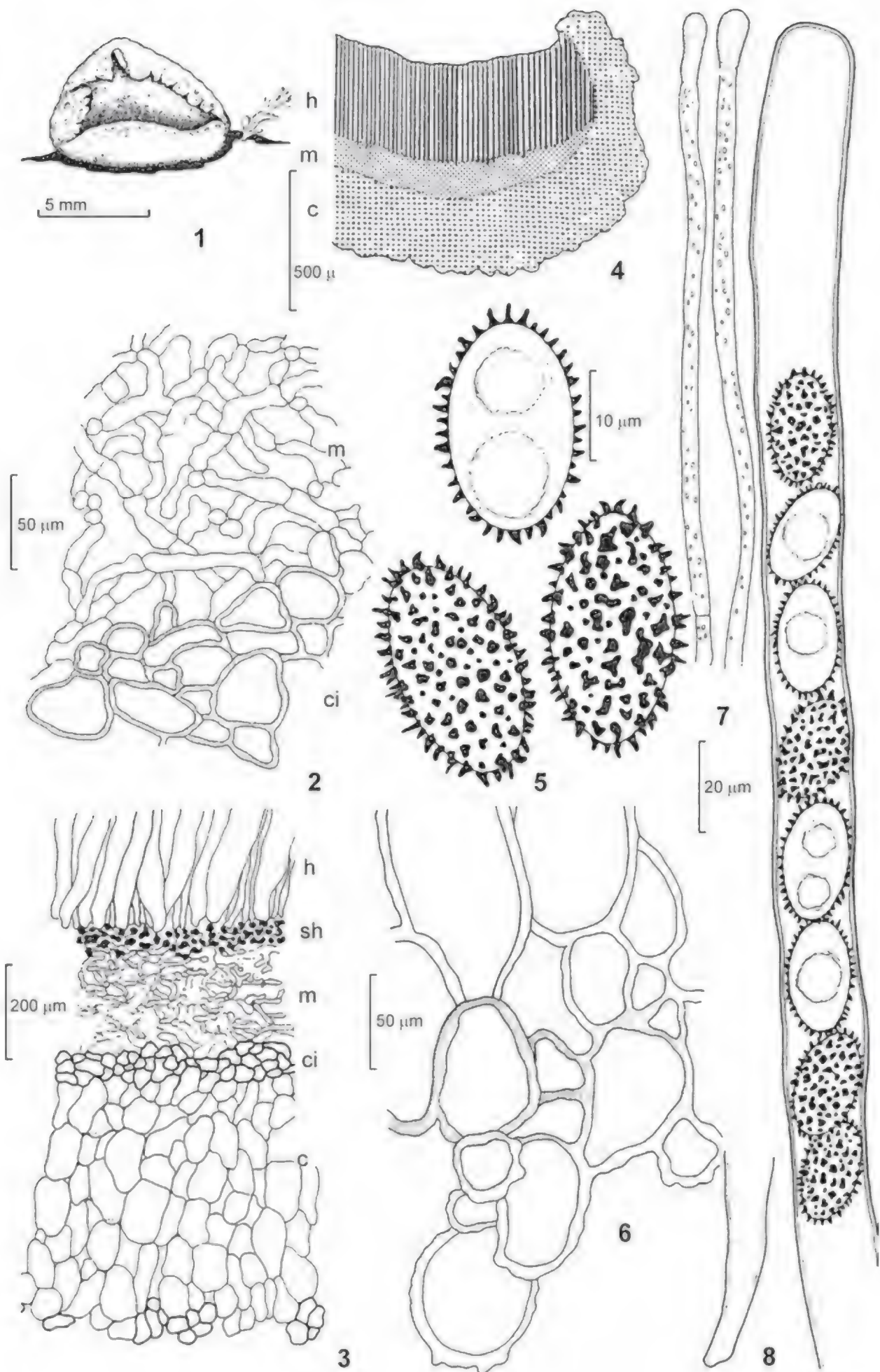
LITERATURE: Eckblad 1968; Dissing 2000; Gamundí 1972a, 1975; Gamundí et al. 2004; Hansen et al. 2001; Korf 1960; 1973a; Rifai 1968; Zhuang & Korf 1986.

### *Anthracobia* Boud. (Pyronemataceae)

ASCOMATA apothecial, small- to medium-sized, sessile, discoid, gregarious of fleshy consistency; disc, smooth, plane to concave, yellowish, ochraceous, orange, reddish to grayish brown; margin conspicuous, undulated, sometimes striated due to tufts of hairs; external surface punctuate, covered irregularly

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PLATE 3. 1–8. *Aleurina echinata* (BAFC 20856). 1. Ascoma. 2. Detail of the medullary excipulum (m) and inner layer of the ectal excipulum (ci). 3. Vertical section of the ascoma: h, hymenium, sh, subhymenium, c, ectal excipulum, m, ci, as in FIG. 2. 4. Sketch of a vertical section of the ascoma: h, m, c, as in FIG. 3. 5. Ascospores. 6. Detail of ectal excipulum. 7. Paraphyses. 8. Ascus.





with bunches of short, superficial, brown, blunt, flexuous or straight hairs with few septa. ECTAL EXCIPULUM of textura angularis composed of isodiametric cells of hyaline to brownish walls, arranged in rows perpendicular to the external surface, the most superficial extending sometimes to form hairs. MEDULLARY EXCIPULUM of textura intricata, hyaline. ASCI cylindrical, 8-spored, J–. PARAPHYSES clavate at the apex and containing granules of pigment. ASCOSPORES uninucleate, 1-seriate, 2- to multiguttulate, sometimes with de Bary bubble, hyaline, smooth, ellipsoidal.

TYPE SPECIES: *Anthracobia melaloma* (Alb. & Schwein.) Arnould, Bull. Soc. Mycol. France 9:112. 1893.

HABITAT: anthracobiontic, typically on burnt soil or charred wood.

ANAMORPH: *Scytalidium*-like, as registered in Kirk et al. (2008). *Scytalidium* Pesante is a dematiaceous hyphomycete that forms chains of brown and hyaline, 0–1 septate arthroconidia.

NOTES: *Anthracobia* is similar to *Melastiza* that also has blunt, brown, short hairs but in this genus the ascospores have ornamented episporium (see description of *Melastiza*). Septal structure in *Anthracobia* ascogenous hyphae (TEM) shows pores filled with an electron-opaque granular matrix, and paraphyses with crystalloid, hexagonal, or rectangular Woronin bodies around the septal pore. It is also related to *Trichophaea*, which has long, pointed hairs. Recent phylogenetic studies based on partial sequences of nLSU rDNA support this relationship but boundaries between both genera remain unclear. It is also suggested that both genera are non-monophyletic. Another molecular study based in SSU rDNA sequences support the close relationship between *Anthracobia* and *Sphaerosporella* (Svrček) Svrček & Kubička, distinct by its globose ascospores. In a recent ecological paper the authors hypothesize that *Anthracobia*, as other postfire fungi, is one of the pivotal species in early restoration of forest systems after disturbance, binding soil particles in the absence of plant roots and potentially helping to reestablish the vegetation.

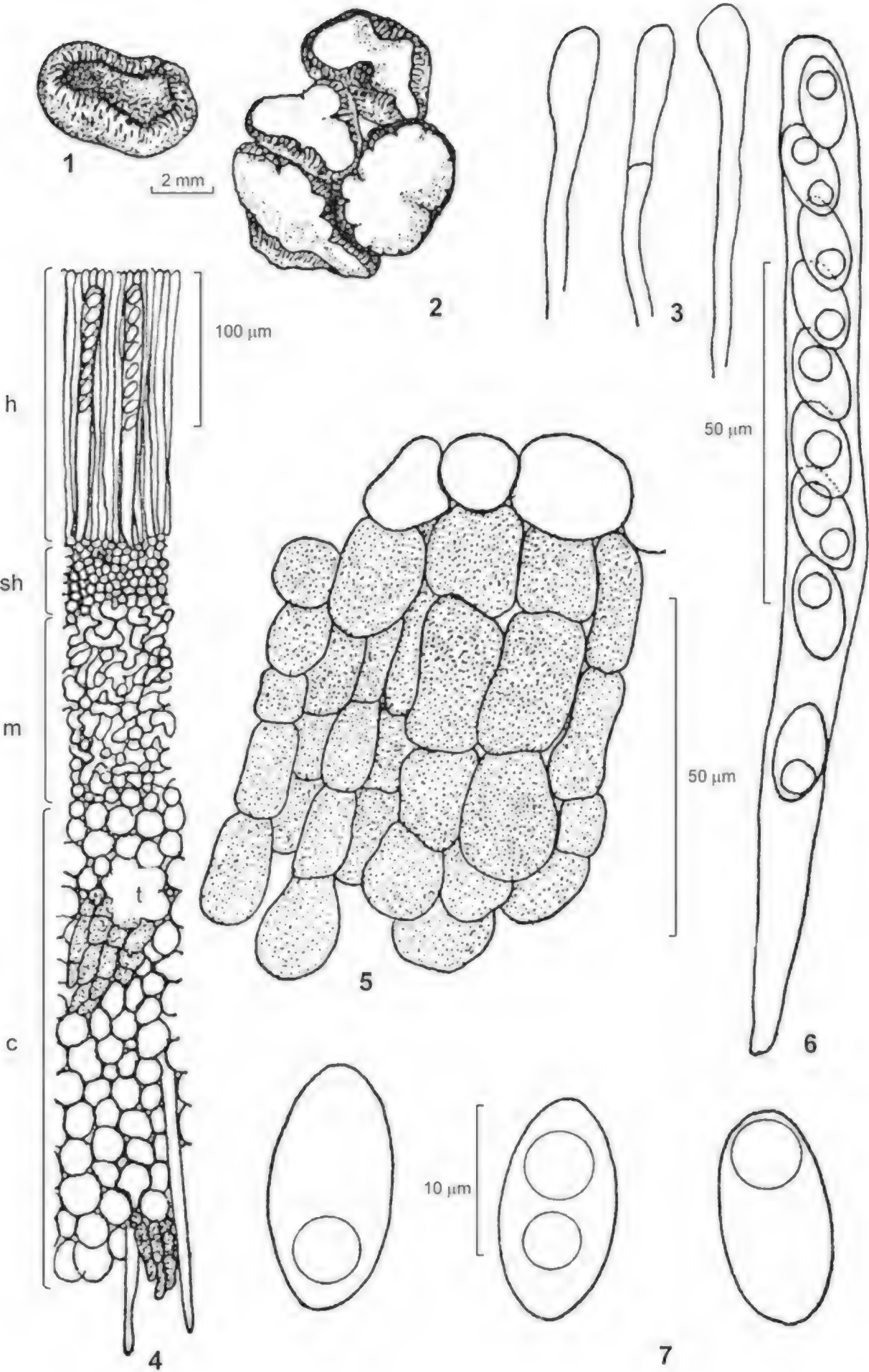
DISTRIBUTION IN ARGENTINA: Two species are recorded: *A. melaloma* and *A. maurilabra* (Cooke) Boud. from BA, ER, S.

ILLUSTRATION: Pl. 4, 1–7. *Anthracobia melaloma*.

LITERATURE: Claridge et al. 2009; Dennis 1978, 1995; Dissing 2000; Gamundí 1960, 1975; Hansen & Pfister 2006; Kimbrough & Curry 1986; Kirk et al. 2008; Liu & Zhuang 2006; Perry et al. 2007; Rifai 1968; Sigler & Carmichael 1976; Sigler & Wang 1990; Svrček & Kubička 1961; Yao & Spooner 1995c, 1996b.

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PLATE 4. 1–7. *Anthracobia melaloma* (LPS 18527). 1. Ascoma. 2. Gregarious ascomata. 3. Paraphyses. 4. Vertical section of the ascoma: h, hymenium, sh, subhymenium, m, medullary excipulum, c, ectal excipulum, t, tuft of hairs. 5. Detail of the ectal excipulum. 6. Ascus. 7. Ascospores.



*Cheilymenia* Boud. (*Pyronemataceae*)

ASCOMATA apothecial small- to medium-sized, superficial, sessile, barrel-shaped, lenticular or scutellate, scattered to gregarious, usually bright coloured; disc smooth, plane to convex, yellow, orange to reddish; margin conspicuous, hairy; external surface the same colour of the disc or paler; hairs hyaline, yellowish to brown, 100–1000  $\mu\text{m}$  long, pluriseptate, simple, forked or bulbous at the base, arising deeply from the excipulum, in some species also with superficial hairs, stellate or hyphoid, mainly at the base of the ascoma. ECTAL EXCIPULUM of textura angularis to textura globulosa of isodiametric cells. MEDULLARY EXCIPULUM poorly differentiated, a textura intricata to epidermoidea of hyaline hyphae. ASCI cylindrical to subcylindrical, operculate, usually 8-spored, J–. PARAPHYSES pluriseptate, straight, subclavate at the apex, usually containing carotenoid granules,  $\gamma$ -carotene as a major pigment. ASCOSPORES uninucleate, 1-seriate, usually eguttulate, hyaline to pale yellowish, ellipsoidal, smooth or verruculose, longitudinally striate, cristulate with crests that can anastomose, the perispore easily separable and delicate, cyanophilic.

TYPE SPECIES: *Cheilymenia stercorea* (Pers.) Boud., Icones Mycol. Liste Préliminaire [3] 1904.

HABITAT: on soil, plant debris, dung, liverworts.

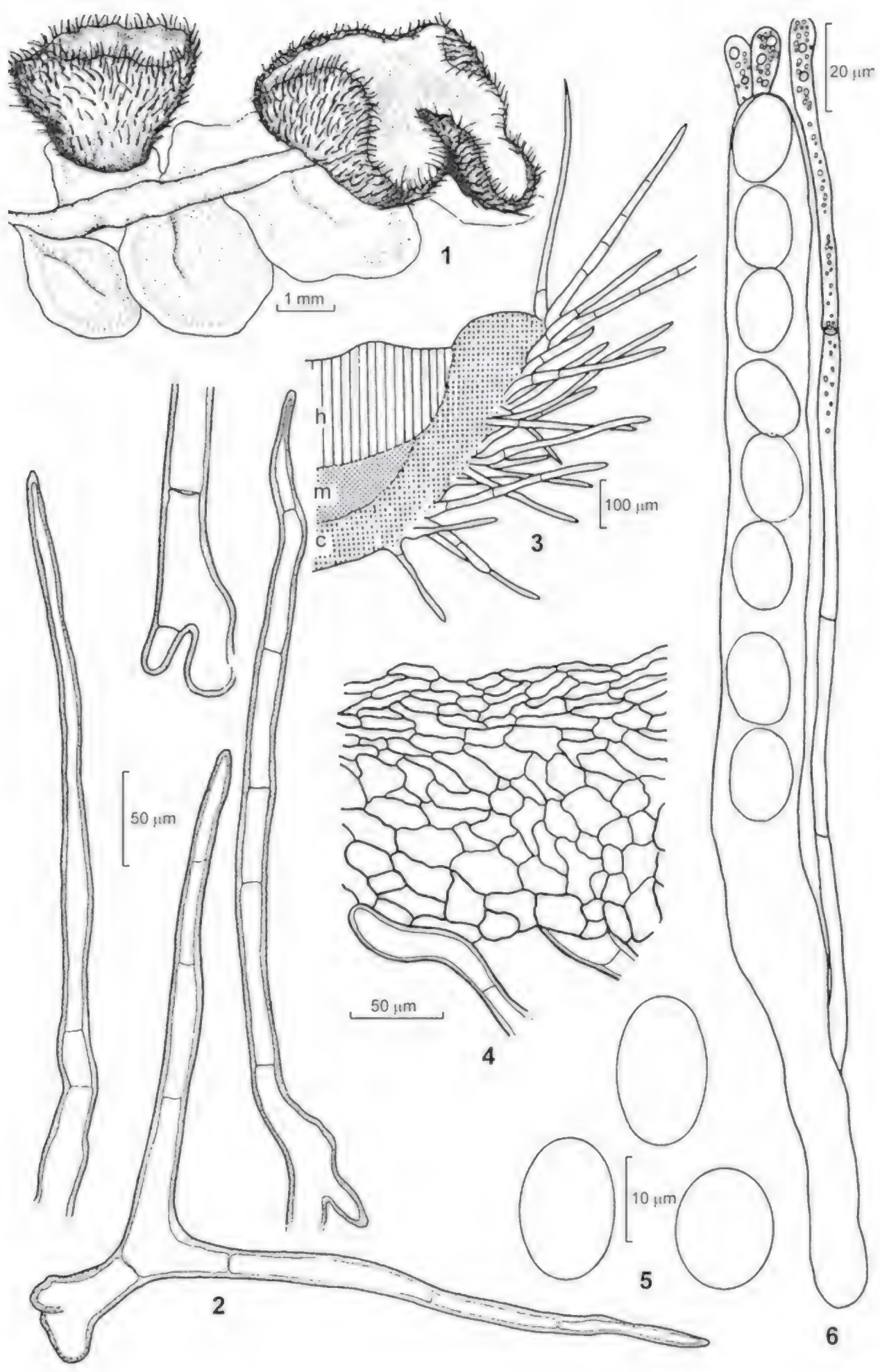
ANAMORPH: unknown.

NOTES: *Cheilymenia* is closely related to *Scutellinia*. Both share morphological characters such as hairs arising deeply from the excipulum and the same major pigment. TEM studies in apothecial tissues of *Cheilymenia* revealed that pores in ascogenous hyphae are occluded by Woronin bodies covered by a deeply staining amorphous electron opaque substance, a scutellinioid-type feature that is shared with *Scutellinia*. A SSU rDNA sequence-based study shows that *C. coprinaria* (Cooke) Boud. [= *C. fimicola* (De Not. & Bagl.) Dennis] and some species of *Scutellinia* are sister groups and that *C. stercorea* is closer to *Byssonectria*. Other studies support the view that this genus is not monophyletic. Several features separate *Cheilymenia* from *Scutellinia*: A) *Cheilymenia* has also superficial hairs and otherwise the rooting hairs can be pigmented or hyaline; B) ascospores eguttulate, if verruculose or striate, show a delicate outer wall (perispore) that separates when treated with lactic acid; and C) globose ascospores have never been recorded. Some species of *Cheilymenia* are related to *Coprobia* but the latter has hairless apothecia, the excipulum totally of textura globulosa, and the robust and capitate paraphyses. However, based on SEM studies in the

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PLATE 5. 1–6. *Cheilymenia villosa* (LPS 36718). 1. Ascomata on liverworts. 2. Marginal hairs. 3. Sketch of a vertical section of the ascoma: h, hymenium, m, medullary excipulum, c, ectal excipulum. 4. Ectal excipulum with basal hairs. 5. Ascospores. 6. Ascus and paraphyses.





ascospore wall, Moravec merged both genera under *Cheilymenia*, proposing an infrageneric classification and recognizing nine sections, including *Coprobiae*. I think that in the sense of this author the concept of the genus is a very wide assemblage of diverse species.

DISTRIBUTION IN ARGENTINA: two species are recorded from a Central province (BA): *C. hyalochaeta* (Speg.) Gamundí and *C. fraudans* (P. Karst.) Boud. and seven species from Patagonia (N, RN, SC, T, TF): *C. megaspora* (Gamundí) J. Moravec, *C. fimicola* *C. humarioides* (Rehm) Gamundí, *C. raripila* (W. Phillips) Dennis, *C. stercorea*, *C. theleboloides* (Alb. & Schwein.) Boud., *C. villosa* Gamundí.

ILLUSTRATION: Pl. 5, 1–6. *Cheilymenia villosa*.

LITERATURE: Arpin 1969; Denison 1964; Gamundí 1960, 1966, 1972b, 1975; Gamundí et al. 2004; Kimbrough 1994; Kimbrough & Curry 1986; Liu & Zhuang 2006; Moravec 1968, 1984, 1989, 1990, 1994b, 1998, 2003, 2005, 2006; Perry et al. 2007; Wu & Kimbrough 1992.

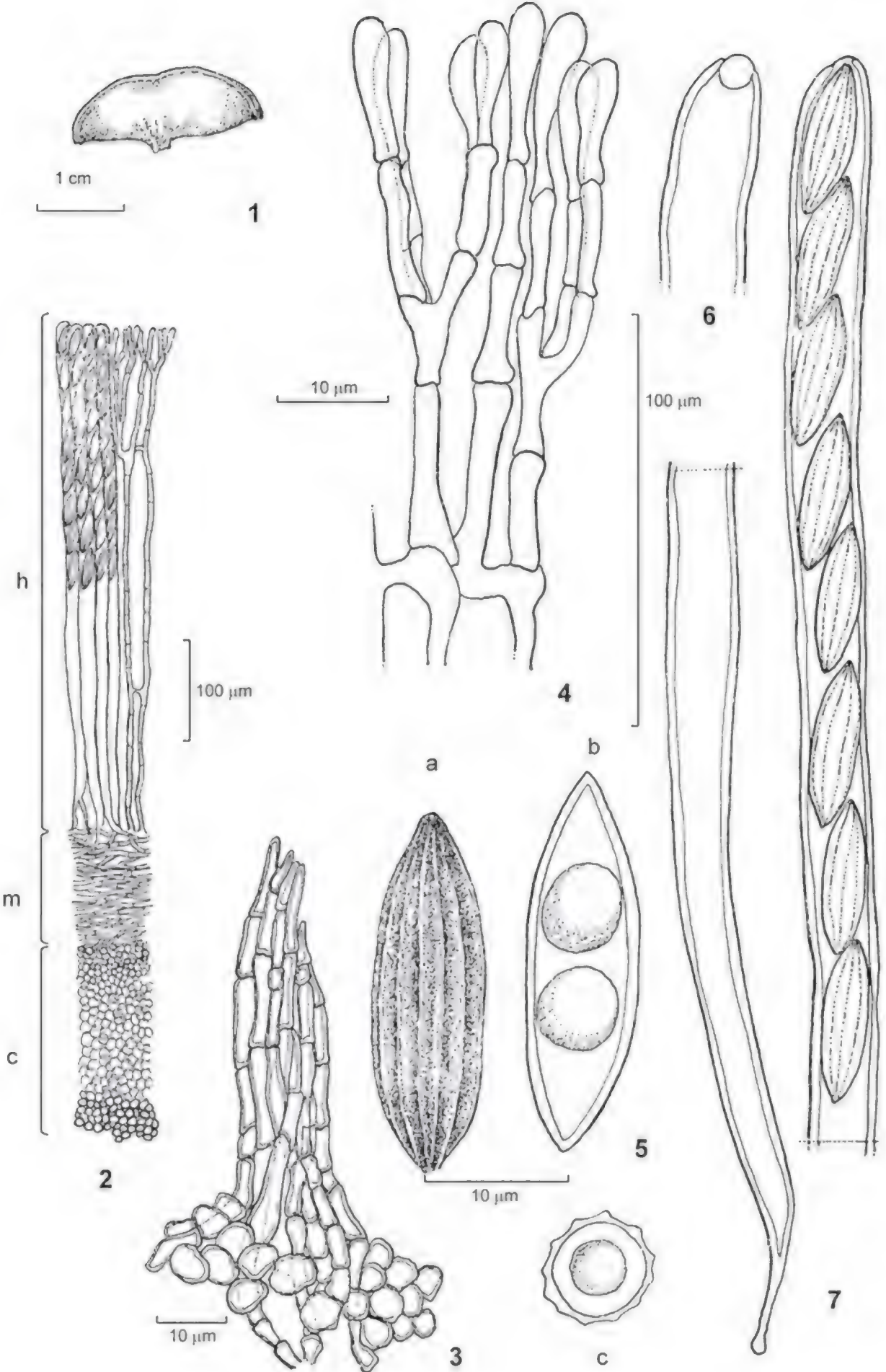
### *Cookeina* Kuntze (*Sarcoscyphaceae*)

ASCOMATA apothecial, medium-sized to large, cup shaped, superficial, sessile, subsessile to stipitate, scattered to gregarious, usually bright coloured, but occasionally pale; disc concave, smooth, in several shades of orange, pink, reddish, purplish or brownish; margin conspicuous, elevated; external surface, pruinose, furfuraceous or hirsute, paler than the disc; furfuration consisting of conical mounds; hairs, when present, fasciculate, covering margin and receptacle, sometimes down the stipe, arising from the medullary or ectal excipulum. ECTAL EXCIPULUM thin, of textura angularis to globulosa of isodiametric cells arranged in rows perpendicular to the surface where they aggregate in conical or dome-like projections that gives the scurfy appearance to the receptacle. MEDULLARY EXCIPULUM well developed, a textura porrecta of hyaline hyphae parallel to the surface of the receptacle, sometimes forming a gelatinous layer, which gives a subgelatinous consistency to the ascoma. ASCI cylindrical to subcylindrical, suboperculate (asymmetrical operculum), thick walled (three layers visible with TEM), contracted below forming an appendiculate base, 8-spored, J–, maturing simultaneously. PARAPHYSES pluriseptate, filiform, straight, profusely branched near the apex, sometimes anastomosing and forming a delicate net, containing carotenoids (major pigment phillipsiaxanthine). ASCOSPORES multinucleate, 1-seriate, containing 1–2 large guttules, hyaline to pale yellowish, ellipsoidal to fusoid, sometimes apiculate at both ends and inaequilateral, smooth or striate, in this case with longitudinal ridges that occasionally anastomose between them or connected

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PLATE 6. 1–7. *Cookeina venezuelae* (LIL, Singer T-2291). 1. Ascoma. 2. Vertical section of the ascoma: h, hymenium, m, medullary excipulum, c, ectal excipulum. 3. Fascicle of excipular hairs. 4. Paraphyses. 5. Ascospores: a, surface view, b, optical vertical section, c, optical cross section. 6. Ascus apex. 7. Ascus.





by fine transverse markings not stained with lactic blue (cyanophobic), wall two layered.

TYPE SPECIES: *Cookeina tricholoma* (Mont.) Kuntze, Revisio Genera Plantarum 2: 849. 1892.

HABITAT: on fallen angiosperm branches, logs, dead twigs or wood and debris.

ANAMORPH: ascospores germinate giving rise to globose to subglobose, hyaline, conidium-like structures.

NOTES: *Cookeina* shows affinities with *Microstoma* Bernstein, *Boedijnopeziza* S. Ito & S. Imai, and *Phillipsia*. *Microstoma* differs in having a multilayered excipulum, simple, flexuous hairs, and universally smooth ascospores. Molecular analysis shows that *Cookeina* and *Microstoma* Bernstein are sister groups. Morphologically *Boedijnopeziza* differs from *Cookeina* by its turbinate or urceolate ascoma and the origin of hairs. Molecular studies demonstrate a close relationship between both genera suggesting synonymy. Therefore the type species, *Boedijnopeziza insititia* (Berk. & M.A. Curtis) S. Ito & S. Imai has been transferred to *Cookeina*. *Phillipsia*, which differs in the microstructure of the ectal excipulum, a textura intricata to porrecta that gives a coriaceous consistency to the ascoma, the simple superficial hairs, and the universally inequilateral ascospores, is widely recognized as different from *Cookeina*.

This genus is pantropical and distributed in the tropics and subtropics. The drawings of *C. venezuelae* (Berk. & M.A. Curtis) Le Gal that illustrate this genus are based on collections from Tucumán, Argentina (LIL T-2291) and show only longitudinal ridges but not the fine transversal interconnecting ridges noted by Iturriaga and Pfister on material from Colombia (FH 1161).

DISTRIBUTION IN ARGENTINA: *C. colensoi* (Berk.) Seaver, *C. tricholoma*, and *C. venezuelae* are recorded from the subtropical area: BA, J, M, T.

ILLUSTRATION: Pl. 6, 1–7. *Cookeina venezuelae*.

LITERATURE: Arpin 1969; Boedijn 1933; Cabello 1988; Denison 1967; Eckblad 1968; Gamundí 1957a, 1959, 1983; Harrington et al. 1999; Iturriaga & Pfister 2006; Le Gal 1953; Meléndez-Howell et al. 2003; Romero & Gamundí 1986; Paden 1975, 1984; Weinstein et al. 2002; Zhuang & Wang 1998.

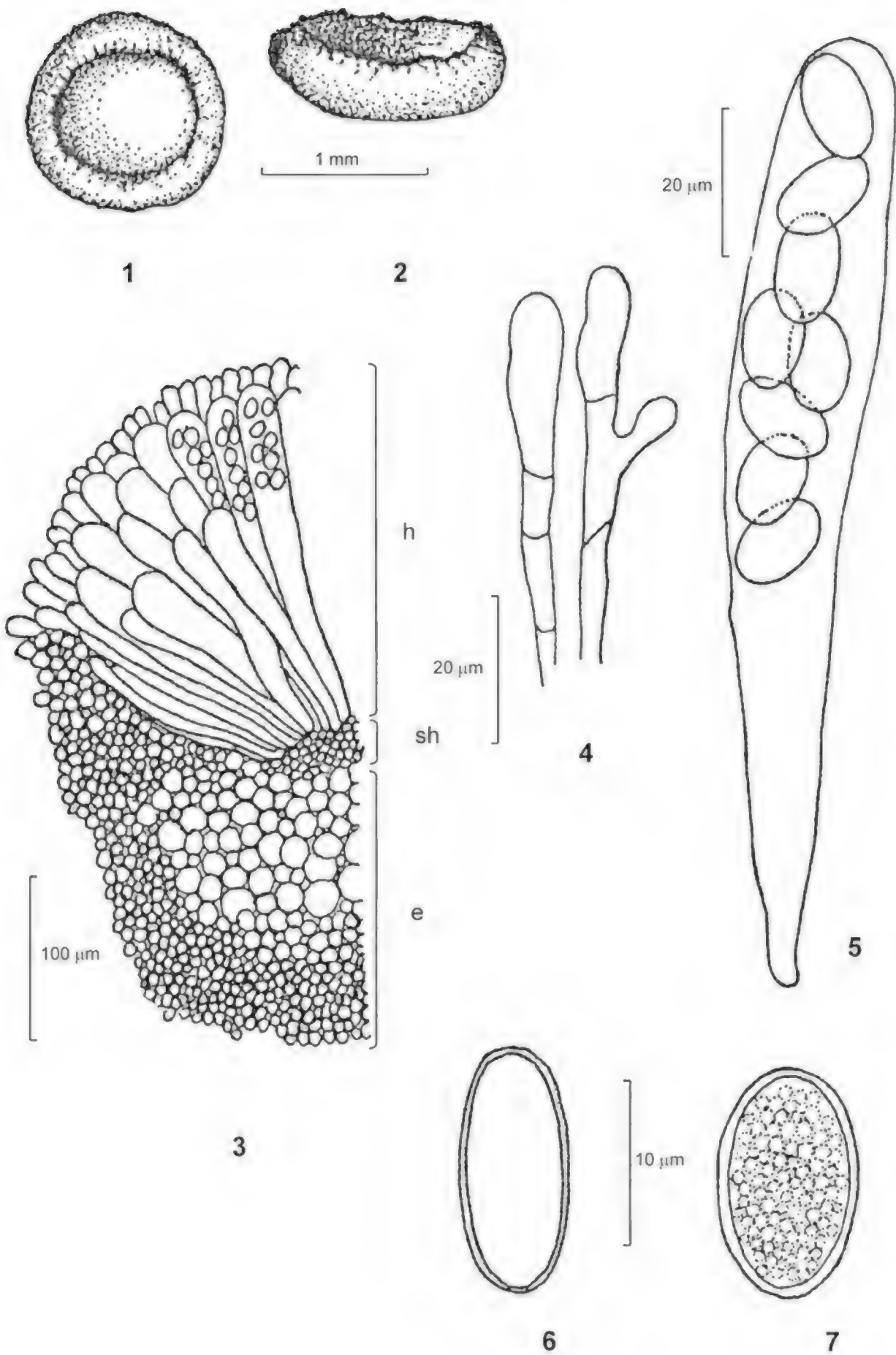
### ***Coprobria* Boud. (*Pyronemataceae*)**

ASCOMATA apothecial, small, superficial, sessile, scutellate to pulvinate, gregarious, ochraceous-orange, disc plane to convex, often granulose due to the protruding ripe asci; external surface hairless, paler than the disc. EXCIPULUM 1-layered, of textura globulosa comprising large, isodiametric cells up to 100 µm,

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PLATE 7. 1–7. *Coprobria granulata* (LPS 27324). 1–2. Ascomata. 3. Vertical section of the ascoma: h, hymenium, sh, subhymenium, e, excipulum. 4. Paraphyses. 5. Ascus. 6. Mature ascospore. 7. Young ascospore.





in the exterior of the receptacle smaller in the inner part. ASCI subcylindrical, operculate, 8-spored, J–. PARAPHYSES pauci-septate, robust, straight, capitate at the apex, containing granules of carotenoids (major pigments  $\beta$ - and  $\gamma$ -carotene). ASCOSPORES uninucleate, 1-seriate, hyaline, without oil guttules, ellipsoidal, smooth or finely striate, with the outer wall easily loosened when heated in lactic acid.

TYPE SPECIES: *Coprobria granulata* (Bull.) Boud., Hist. class. Discom. d'Europe: 69. 1907.

HABITAT: on dung of several herbivorous mammals and manure.

ANAMORPH: unknown.

NOTES: *Coprobria* is related to *Cheilymenia*. Main distinctions are: A) the latter is conspicuously hairy, the hairs being rooting and sometimes having superficial hairs; B) the excipulum usually differentiated in two layers; and c) paraphyses are more slender than in *Coprobria*. Moravec at first recognized *Coprobria*, including also a new species, but in his later revision of *Cheilymenia*, he considered *Coprobria* a section of *Cheilymenia* (see NOTES under *Cheilymenia*). His conception of this genus is very wide and is based mainly on the morphology of the ascospores. Other authors maintain *Coprobria* as a separate genus, emphasizing the structure of the excipulum, entirely of textura globulosa, and the receptacle devoid of hairs. These features are nowadays accepted in discomycete taxonomy as valuable characters for distinguishing genera. I accept this view. Furthermore, in *Coprobria granulata* the hymenial surface appears rough from protruding asci and capitate paraphysis, a character absent in *Cheilymenia*.

DISTRIBUTION IN ARGENTINA: only *C. granulata* is recorded from BA. Rehm (1899) described *Humaria granulata* f. *guanacensis* Rehm and *Humaria guanaci* Rehm from Tierra del Fuego, both on "guanaco" (*Lama guanicoe*) dung. These most probably are *Coprobria granulata*, but the type specimens are both missing.

ILLUSTRATION: Pl. 7, 1–7. *Coprobria granulata*.

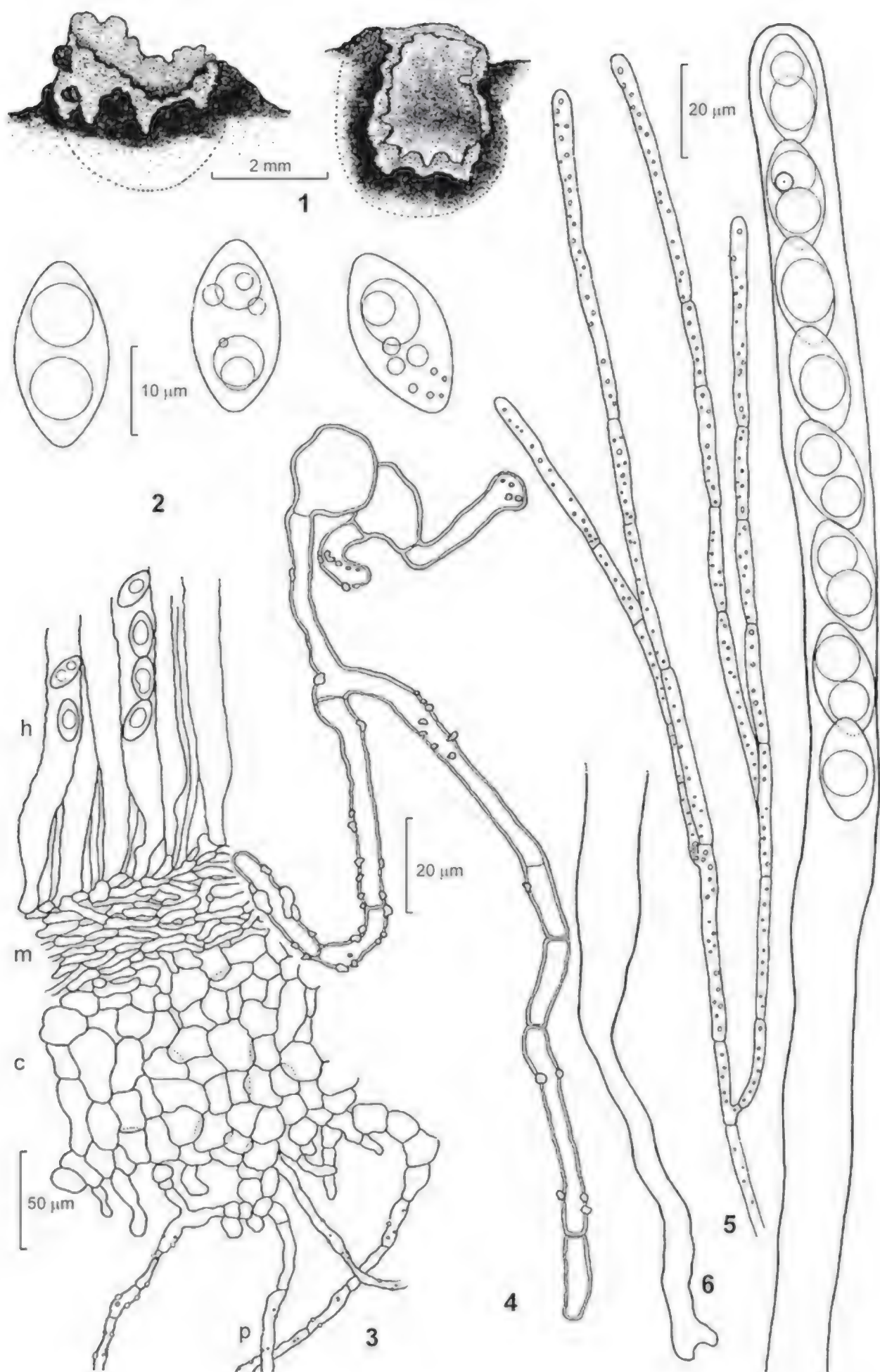
LITERATURE: Arpin 1968; Dennis 1978, 1986, 1995; Gamundí 1960, 1975; Gamundí et al. 2004; Moravec 1984; Rifai 1968; Rehm 1899.

### *Geopora* Harkn. emend. Burds. (Pyronemataceae)

ASCOMATA cupulate, globose to subglobose, solitary or gregarious; hypogeous forms closed but sometimes opening superficially at maturity, discharging ascospores actively (puffing); hymenial surface concave or convoluted; outer surface covered by a dense mat of dark hairs. Hymenium smooth or convoluted (ptychothecia). ECTAL EXCIPULUM of textura angularis, cells up to 60  $\mu$ m diam.,

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PLATE 8. 1–6. *Geopora arenicola* (BAFC 21685). 1. Ascomata: lateral and frontal view. 2. Ascospores. 3. Vertical section of the ascoma: h, hymenium, m, medullary excipulum, c, ectal excipulum, p, hairs. 4. Detail of hairs. 5. Paraphyses. 6. Ascus base and upper portion.





thick walled, dark, giving rise to superficial, flexuous, pigmented hairs, ending in obtuse tip (8–14  $\mu\text{m}$  thick), simple or branched, multiseptate. MEDULLARY EXCIPULUM a textura intricata of hyaline, thin walled hyphae. ASCI cylindrical, operculate, 8-spored, J–, arranged in a hymenial layer. PARAPHYSES slender, thin walled, septate, hyaline or slightly swollen at the apex. ASCOSPORES uninucleate, uniseriate, 1- or multiguttulate, containing oil droplets, hyaline, thin-walled, smooth, subglobose to elliptical, sometimes collapsing laterally.

TYPE SPECIES: *Geopora cooperi* Harkn., Bull. California Acad. Sci. 1: 168. 1885.

HABITAT: in or on soil, under various species of trees or shrubs.

ANAMORPH: unknown.

NOTES: *Geopora* is here considered in the concept of Burdsall, which contains not only the hypogeous but also the epigeous species included by Boudier in *Sepultaria* (Cooke) Boud. The type species, *G. cooperi*, may be hypogeous as well as epigeous. The position of the ascoma regarding the soil surface, which appears to have evolved independently multiple times within the *Pyronemataceae*, is not considered diagnostic for the genus or phylogenetically significant. Formerly placed in *Tuberales*, *Geopora* was moved by Burdsall to the *Pezizales* (*Pyronemataceae*) because the asci are operculate. The genus is related to *Hydnocystis* Tul. & C Tul. (formerly *Tuberales*, now also *Pezizales*), which has asci without operculum and paraphyses forming an epithecium. *Geopora* is also related to *Trichophaea* (*Pezizales*, *Pyronemataceae*), which differs in possessing rigid hairs. A molecular study demonstrated that some *Geopora* species are mycobionts forming ectomycorrhiza with coniferous and deciduous trees. The corresponding phylogenetic analysis suggests affinities with *Tricharina*.

DISTRIBUTION IN ARGENTINA: widely distributed in the Northern Hemisphere, only one species is recorded: *Geopora arenicola* (Lév.) Kers, cited as *Sepultaria arenicola* (Lév.) Massee from BA, ME, RN, TF.

ILLUSTRATION: Pl. 8, 1–6. *Geopora arenicola*.

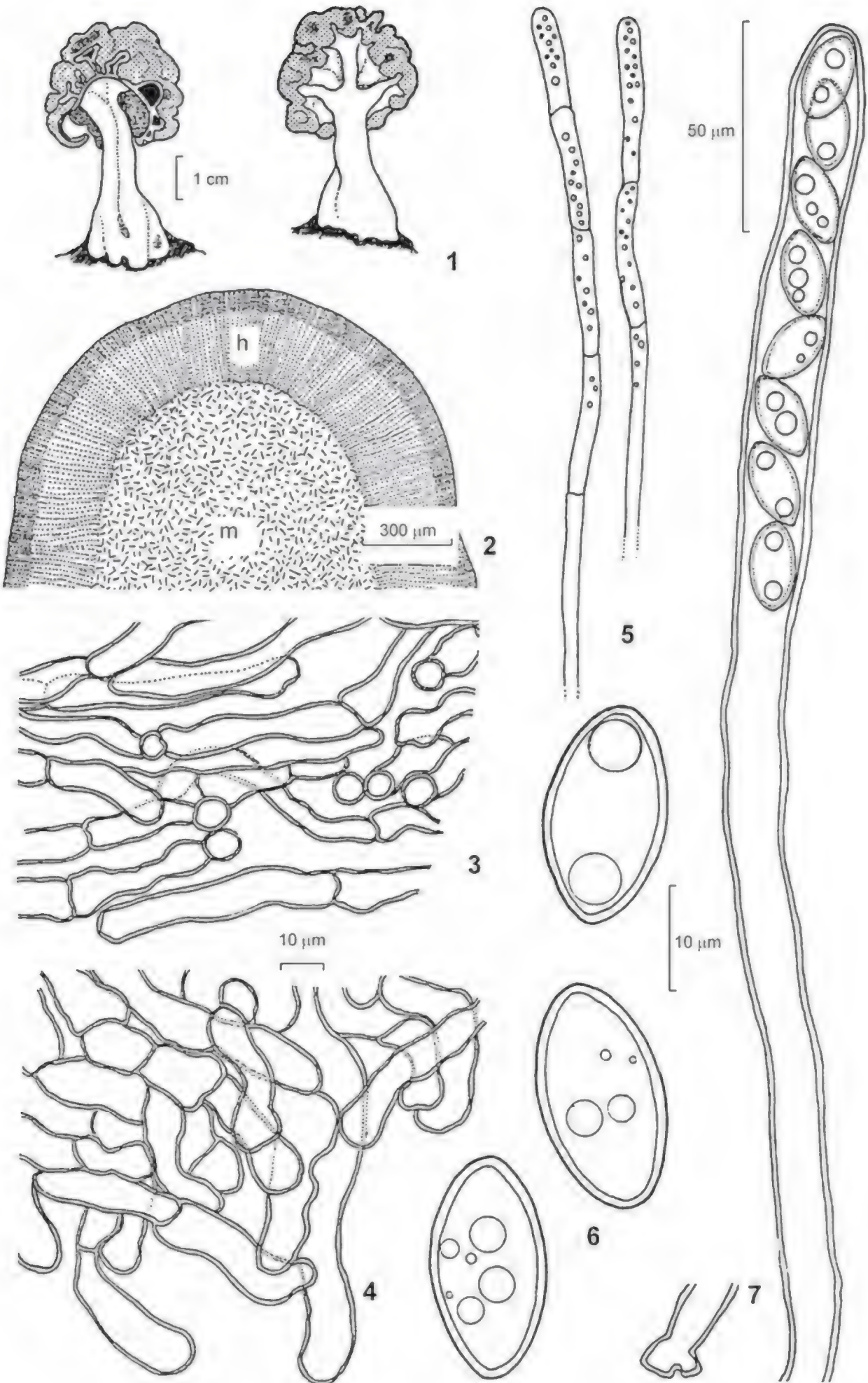
LITERATURE: Boudier 1885; Burdsall 1968; Gamundí 1960, 1975; Korf 1973a; Læssøe & Hansen 2007; Tedersoo et al. 2006; Trappe 1979; Yang & Korf 1985a,b; Yao & Spooner 1996a; Zhang & Yu 1992.

### ***Gyromitra* Fr., nom. cons. (*Discinaceae*)**

ASCOMATA apothecial, cupulate, discoid, convex to undulate or pileate with pileus irregularly lobed or convoluted, medium-sized to large; superficial, sessile to distinctly stipitate; solitary or scattered; fleshy consistency, leathery when dry; hymenium yellow-brown, orange-brown, chesnut-brown

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PLATE 9. 1–7. *Gyromitra antarctica* (BAFC 22009). 1. Ascomata. 2. Sketch of a cross section of the pileus: h, hymenium, m, medullary excipulum. 3. Detail of medullary excipulum (m, in FIG. 2). 4. External hyphae from the stipe. 5. Paraphyses. 6. Ascospores. 7. Ascus base and upper portion.



to dark brown; margin free, recurved; external surface glabrous to pubescent, paler than the hymenium; stipe cylindrical, terete or slightly sulcate, tapering or bulbous near the base, solid, hollow or lacunose, white or with a reddish tinge at the base, glabrous to pubescent. EXCIPULUM entirely of textura intricata at maturity, composed of hyaline, inflated hyphae with septal structure (TEM) showing pores occluded by an electron-dense material and surrounded by elongate Woronin bodies. ASCI cylindrical to subcylindrical, 8-spored, J-. PARAPHYSES straight or forked, robust, slightly enlarged at the apex, with an extracellular incrustated and/or intracellular diffuse reddish brown pigment. ASCOSPORES 4-nucleate, 1–2 seriate, usually containing 1–2 lipid guttules, if 3 the central one larger, hyaline, ellipsoidal to subfusoidal, with or without apiculi at both poles, smooth, verruculose to finely reticulate (SEM), with cyanophilic perispore.

TYPE SPECIES: *Gyromitra esculenta* (Pers.) Fr. Summa Veg. Scand., Pars Posterior: 346. 1849.

HABITAT: on soil in deciduous or coniferous forests along path and disturbed areas or on decaying wood, in springtime.

ANAMORPH: unknown.

NOTES: *Gyromitra* is taken here in the wide sense of Harmaja to include *Discina* (Fr.) Fr., *Paradiscina* Benedix, and *Neogyromitra* S. Imai; the first is considered a subgenus distinguished by subsessile, convex ascomata and 3-guttulate, apiculate ascospores, features that Harmaja considered of quantitative value, since the excipulum and ascospore wall structures are similar. TEM study of the origin of ascospore walls in *Gyromitra* showed that apiculum and/or spore wall arise from blebbing of the primary wall material through the episporium into the secondary wall upon which a fibrillar deposit from the perispore sac forms the ornamentation. It shares with *Helvella* 4-nucleate ascospores with lipid guttules and the septal ultrastructure, but the pileus and excipular structures differ. (See description of *Helvella*). Some species of *Gyromitra* are poisonous due to a heat-labile substance called gyromitrin. *Gyromitra* (false morel or lorchel) also shows affinity with *Morchella* (known commonly as morel, a precious edible mushroom) because of the pileate ascomata. Both coexist in similar habitats in springtime in SW Argentina. The collector should recognize the latter by the typically honeycombed ochre or grey-brown pileus.

DISTRIBUTION IN ARGENTINA: only one species, *G. antarctica* Rehm, is recorded with certainty from the Andean-Patagonian forest from N, RN.

ILLUSTRATION: Pl. 9, 1–7. *Gyromitra antarctica*.

LITERATURE: Abbott & Currah 1997; Benedix 1966, 1969; Eckblad 1968; Gamundí 1960, 1971; Gamundí & Horak 2003; Gamundí et al. 2004; Harmaja 1973, 1976a,b; Häffner 1987; Kimbrough 1994; Kimbrough et al. 1990; Kimbrough & Gibson 1991.



*Helvella* L. (Helvellaceae)

ASCOMATA epigeous, cupulate, auriculoid or pileate with a pileus discoid, saddle-shaped or lobate, rarely sparassoid, small to large, superficial, sessile to stipitate, solitary, scattered or gregarious, of fleshy consistency; hymenium (disc) whitish to cream coloured, grayish, brown to black; margin free, involute to recurved, undulate, entire to crenate, sometimes with crystalline deposits; external surface glabrous, pubescent to villose, concolorous or paler than the disc; stipe cylindrical, terete or externally sulcate, with longitudinal ribs that may anastomose and invade the receptacle, solid, hollow or lacunose, white, cream coloured or pale grayish to dark gray-brown, glabrous, pubescent to villose. ECTAL EXCIPULUM of textura prismatica to angularis, composed of doliform cells arranged in rows perpendicular to the surface, the outermost clavate, hyaline or with brownish walls, or aggregated in fascicles. MEDULLARY EXCIPULUM a textura intricata of hyaline, branched hyphae, mostly loosely arranged. ASCI cylindrical tapered to the base, aporhynchous or pleurorhynchous, 8-spored, J–. PARAPHYSES straight, cylindrical or slightly enlarged at the apex, hyaline or containing dark brown pigment, pluriseptate. ASCOSPORES 4-nucleate, 1-seriate, usually containing one large lipid guttule, hyaline, broad ellipsoidal to subfusoidal, smooth to verruculose. STIPE in cross section, when it is hollow, shows an extra inner layer like the ectal excipulum.

TYPE SPECIES: *Helvella crispa* (Scop.) Fr., Systema Mycologicum 2(1): 14. 1822.

HABITAT: on damp sandy, clayish, or rich soils, along paths in deciduous and coniferous forests or arctic-alpine vegetation, occasionally on decaying wood.

ANAMORPH: unknown.

NOTES: As a very old name, *Helvella* (or *Elvela*) has been subject to different interpretations and typifications. Modern authors concur in the selection of the type species as presented here. The genus is widely distributed in the Northern Hemisphere and only occasionally collected in Argentina in sites planted with boreal trees. Phylogenetic classifications propose to place it in the family Helvellaceae, which includes not only epigeous genera such as *Helvella* but also the related hypogeous *Barssia* Gilkey and *Balsamia* Vittad., formerly placed in the Tuberaceae. *Helvella* shares with *Gyromitra* the 4-nucleate ascospores, the same spore wall ontogeny, lipid guttules, and the septal ultrastructure of excipular hyphae (TEM), except that *Helvella* usually has spherical Woronin bodies (see description of *Gyromitra*). *Helvella* is related to *Underwoodia*, which has distinctive ascoma morphology with a pileus completely adnate to stipe and coarsely ornamented ascospores. Earlier authors suggested a synonymy with *Helvella*, but various molecular studies support *Underwoodia* as a separate genus (see also *Underwoodia*). *Wynnella* Boud. is currently considered a synonym of *Helvella*. *Pindara* Vel. has been merged with *Helvella* supported

by molecular studies. Several species of *Helvella* have ectomycorrhizal lifestyle. Molecular analyses indicate that they are mycobionts of *Quercus robur* and *Fagus sylvatica*.

DISTRIBUTION IN ARGENTINA: two species, *H. leucomelaena* (Pers.) Nannf. (= *Acetabula nemoralis* Speg.) and *H. leucopus* Pers. have been recorded from BA and N, in gardens and parks.

ILLUSTRATION: Pl. 10, 1–9. *Helvella leucopus*.

LITERATURE: Abbott & Currah 1988, 1997; Berthet 1964; Dissing 1966; Gamundí 1960; Gamundí & Giaioti 1998; Harmaja 1974; Häffner 1987; Hansen & Pfister 2006; Kimbrough 1991, 1994; Kimbrough & Gibson 1990; Korf 1973a; Landvik et al. 1999; Nannfeldt 1937; O'Donnell et al. 1997; Tedersoo et al. 2006.

### *Melastiza* Boud. (*Pyronemataceae*)

ASCOMATA apothecial, medium-sized to large, superficial, sessile, scutellate to cupuliform, scattered to gregarious, bright coloured, disc smooth, plane to concave, orange to red; margin conspicuous, entire or undulate, pruinose to furfuraceous; external surface concolorous with the disc, paler at the base. ECTAL EXCIPULUMA a textura globulosa to angularis comprising isodiametric cells, smaller towards the surface, sometimes brownish; hairs disposed in tufts, giving the margin and the receptacle a pruinose appearance, short, obtuse, brown-walled, with few septa, arising from superficial cells. MEDULLARY EXCIPULUM a textura intricata of densely arranged hyaline hyphae. ASCI cylindrical, 8-spored, J– PARAPHYSES pluriseptate, subclavate or bent at the apex, containing granules of carotenoid pigments ( $\beta$ - and  $\gamma$ - carotene, ester of aleuriaxanthine) that turned green with iodine. ASCOSPORES 1-seriate, uninucleate, 1–2 guttulate, hyaline to pale yellowish, ellipsoidal, with a conspicuous cyanophilic net-like ornamentation, with spiny or hood-like projections at both ends, or with coarse, irregular warts.

TYPE SPECIES: *Melastiza miniata* (Fuckel) Boud., Icon. Mycol. Liste Sér. 1 [3].

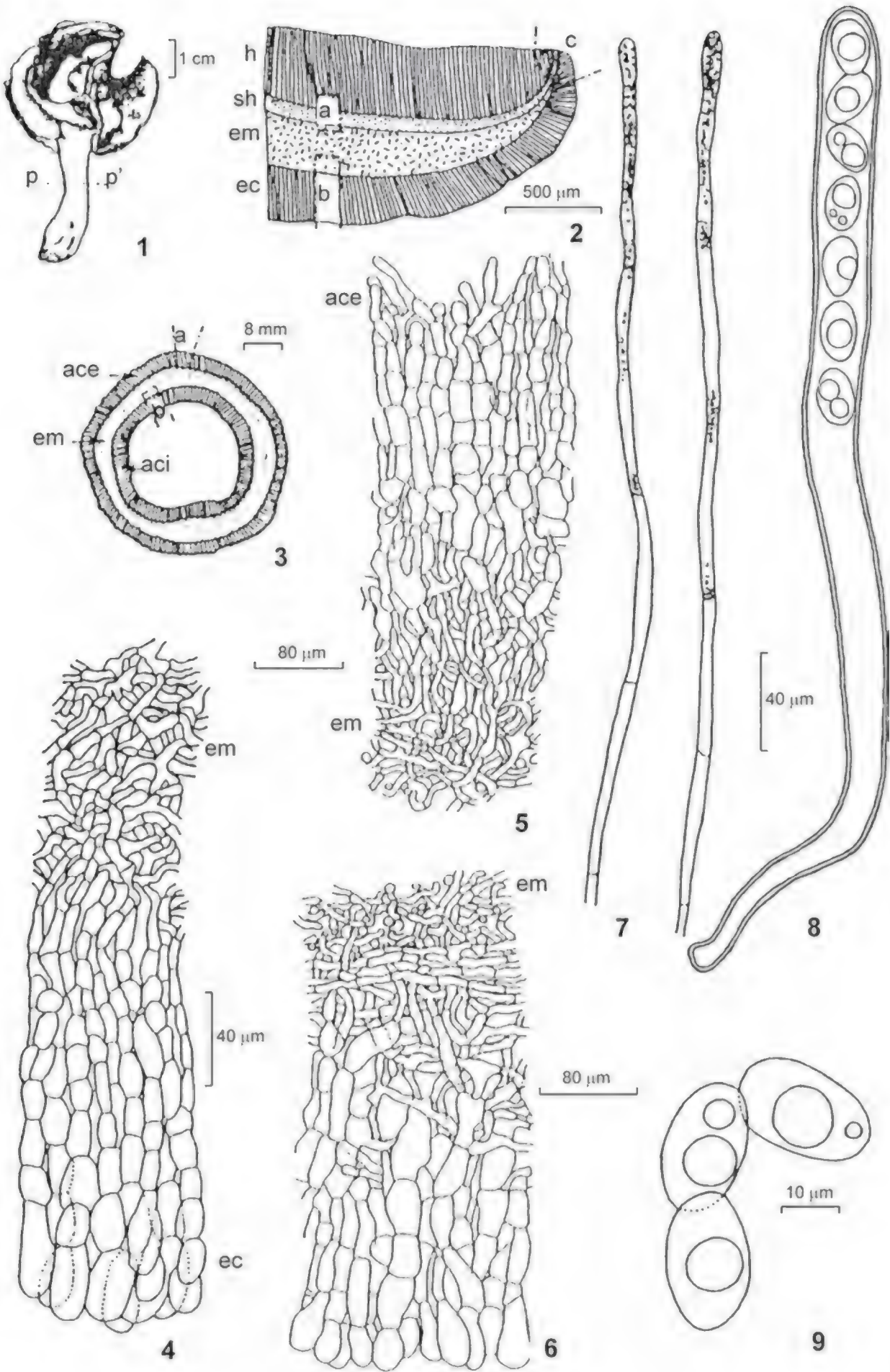
HABITAT: on damp, bare, or sandy soil, sometimes among mosses or on burnt places.

ANAMORPH: unknown.

NOTES: According to Korf (1985) the correct name for *M. miniata*, type species of the genus, is *M. cornubiensis* (Berk. & Br.) J. Moravec. *Melastiza chateri* (W.G. Sm.) Boud.) is often found in association with *Aleuria aurantia* in damp and mossy places but can be distinguished easily by the margin and external surface

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PLATE 10. 1–9. *Helvella leucopus* (BCRU 1489). 1. Ascoma. 2. Sketch of a vertical section of the pileus: h, hymenium, sh, subhymenium, em, medullary excipulum, ec, ectal excipulum. 3. Sketch of a cross section of the stipe (p–p' in FIG. 1)): aci, internal ectal excipulum, em, medullary excipulum, ace, external ectal excipulum. 4. Vertical section of the pileus: detail of b in FIG. 2. 5–6. Detail of a cross section of the stipe (a, b in FIG. 3). 7. Paraphyses. 8. Ascus. 9. Ascospores.





of the ascoma. *Melastiza* is close to *Aleuria* (see NOTES under *Aleuria*), as was pointed out by various authors. Moravec united both genera, placing *Melastiza* as a subgenus of *Aleuria*. His viewpoint is supported by: a) the same type of ornamentation; b) the same carotenoid composition in paraphyses ( $\beta$ - and derivatives of  $\gamma$ -carotene); and c) the same habitat. He neglected the difference concerning hairy (*Melastiza*) vs. hairless (*Aleuria*) ascomata. The hairy feature and hair morphology is often considered important in distinguishing genera within the *Pezizales* (see treatment of *Cheilymenia* and *Coprobria*), so that some authors accept *Melastiza* as a good genus. We adhere to the view that only species with superficial, dark, blunt hairs belongs to *Melastiza* and others with acuminate hairs should be excluded.

DISTRIBUTION IN ARGENTINA: only *M. chateri* is recorded from TF.

ILLUSTRATION: Pl. 11, 1–9. *Melastiza chateri*.

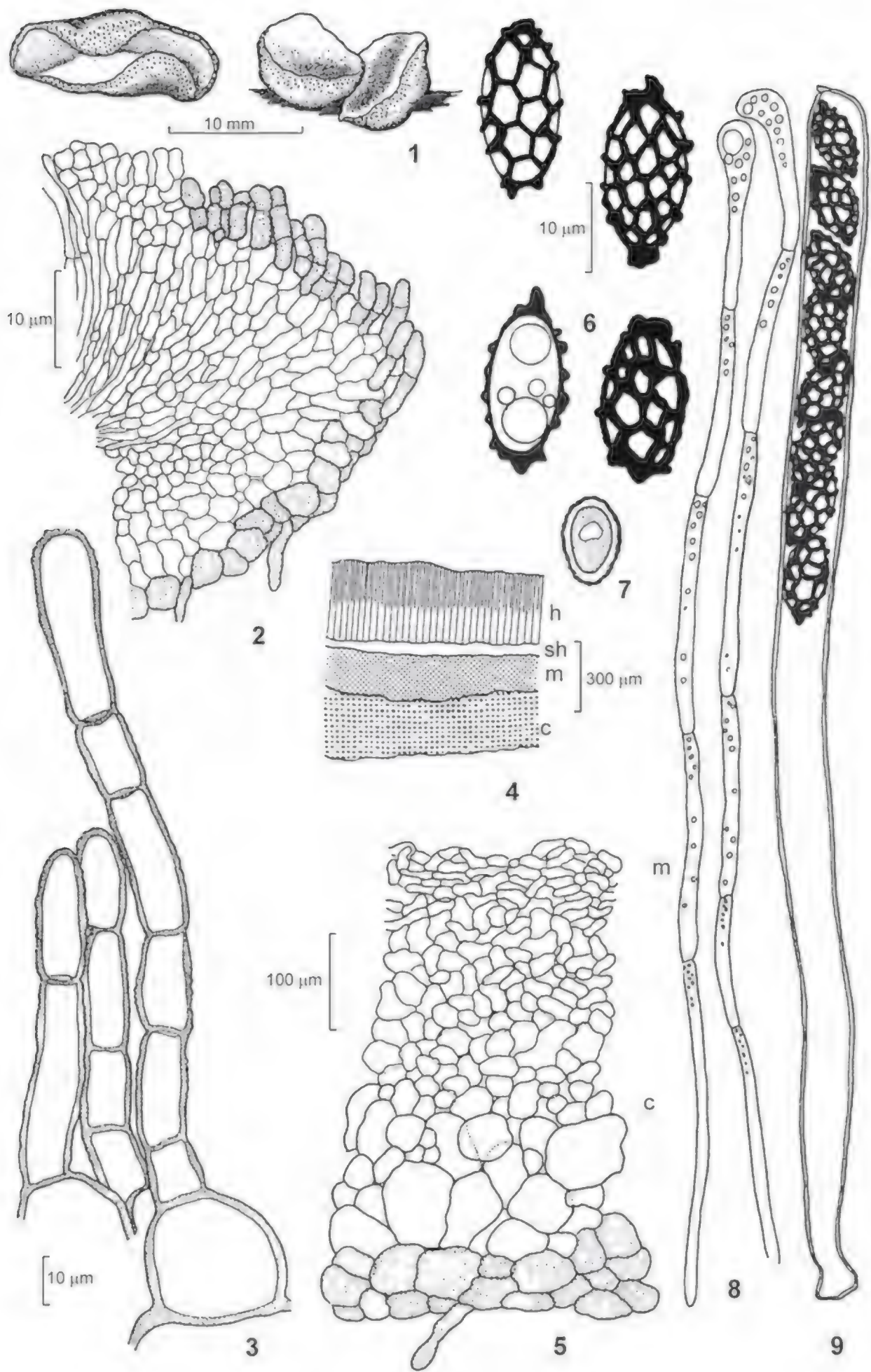
LITERATURE: Arpin 1968; Arroyo & Calonge 1988; Dennis 1978, 1986, 1995; Dissing 2000; Gamundí 1975; Gamundí et al. 2004; Korf 1985; Lassueur 1980; Le Gal 1958; Maas Geesteranus 1967; Moravec 1972, 1994a; Rifai 1968; Yao & Spooner 1995a,b.

### *Morchella* Dill. ex Pers. (*Morchellaceae*)

ASCOMATA pileate, stipitate, large, up to 30 cm high, superficial, gregarious, of fleshy to paperaceous consistency; pileus conical, ovoid to globose, alveolate, alveolae isodiametric or elongate, separated by sterile ribs, giving a honeycomb-like aspect, adnate or separated from the stipe by a shallow groove, or in some species a deep groove; hymenium covering the alveola, ochre, yellow-brown, yellow-orange or grayish-brown, primary ribs concolorous or darker than the hymenium, sterile, longitudinal and anastomosing, sometimes connected with secondary transverse ribs covered by the hymenium; stipe cylindrical or slightly furrowed, bulbous or tapering to the base, hollow, externally glabrous, furfuraceous or scaly, usually whitish or cream, always paler than the pileus. MEDULLARY EXCIPULUM of textura angularis, hyaline. STIPE comprising a cortical layer of textura globulosa to angularis composed of hyaline cells, the most external cylindrical, aggregated in tufts to form furfurations or scales and a inner layer of textura intricata, hyaline. ASCI cylindrical, 8-spored, thin walled, J–. PARAPHYSES robust, straight or curved, capitate, clavate or irregularly enlarged at the apex, diffusely pigmented. ASCOSPORES 1-seriate, multinucleate, at maturity eguttulate, after puffing with external, polar guttules, hyaline to subhyaline, ellipsoidal to subfusoidal, smooth or with delicate longitudinal striation (SEM). Spore print yellowish to orange-pinkish.

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PLATE 11. 1–9. *Melastiza chateri* (BAFC 21982). 1. Ascomata. 2. Marginal excipulum. 3. Detail of hairs. 4. Sketch of a vertical section of the ascoma: h, hymenium, sh, subhymenium, m, medullary excipulum, c, ectal excipulum. 5. Detail of the excipulum: m and c, as in FIG. 4. 6. Mature ascospores in surface view and optical section. 7. Immature ascospore. 8. Paraphyses. 9. Ascus.



TYPE SPECIES: *Morchella esculenta* (L.) Pers., Syn. Meth. fung. 2: 618. 1801.

HABITAT: on calcareous or sandy soils in conifer/deciduous forests or in nearby prairies, disturbed places mixed with charcoal, in orchards or gardens. Saprotrophs and ectomycorrhizal.

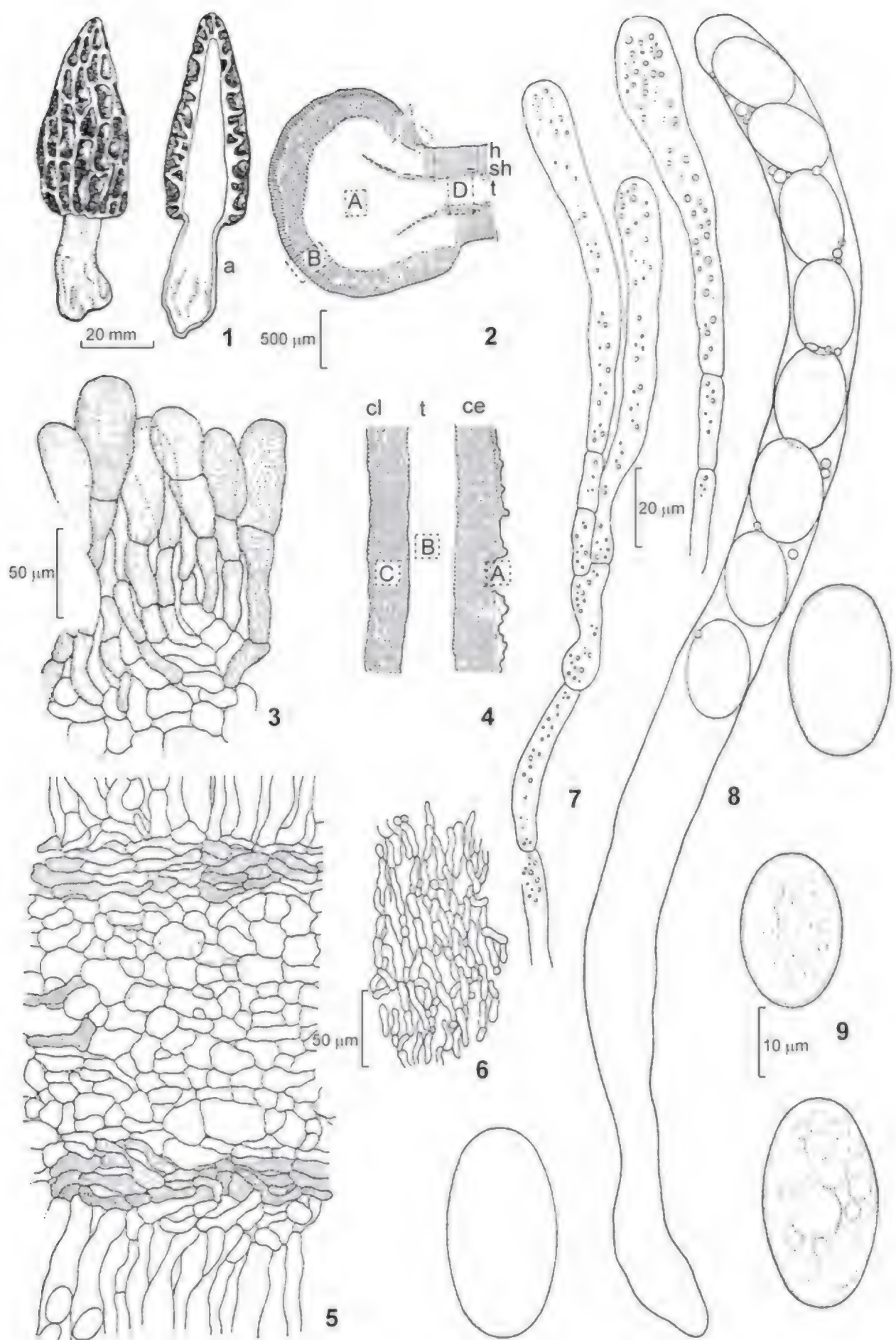
ANAMORPH: *Costantinella* Matr. Conidiophores mononematose, hyaline, conidiogenous cells disposed in whorls, strongly recurved, denticulate; conidia hyaline, sympodioblastic, globose to subglobose.

NOTES: *Morchella* as here understood includes *Mitrophora* Lév., which differs only in having a deep groove separating the cap from the stipe. As a genus *Morchella* is easily identified by its honeycombed pileus but difficult to define due to the uniformity of microscopic features and the variation of morphotypes in nature. In their enzyme-linked immunoabsorbent assay of *Morchella esculenta* complex, for instance, Jung et al. (1993) concluded that immunologically distinguishable forms do produce easily distinguishable morphotypes (e.g. *M. esculenta*). One proposed classification considers three sections (*Adnatae*, *Semiadnatae*, *Distantes*) based on the macroscopic separation of pileus and stipe. *Morchella* shares a pileate ascoma with *Helvella* but differs in the microstructure of the pileus and multinucleate ascospores (see description of *Helvella*). Different also is *Verpa* Sw., which has a campanulate, longitudinally furrowed or reticulate pileus with a free margin and 2–8 ascospores. Molecular generated phylogenies suggest that *Morchella*, *Verpa*, and *Disciotis* Boud. comprise a clade that is sister to *Gyromitra*–*Hydnотrya*. Some species form sclerotia in nature and in vitro. Ectomycorrhizae have been demonstrated between some species of *Morchella* and conifers (*Abies*, *Picea*, *Pinus*). It has been suggested that in nature *Morchella* spp. follow two ecological strategies — either pioneer saprotrophs and ephemeres on disturbed soils or perennial ectomycorrhizals with vascular plants in forests. In both cases they form sclerotia in winter. Fructification in vitro was first reported by Ower and registered as a US Patent. Later a life cycle could be reproduced from ascospores to ascomata suggesting two alternate pathways, via a) primary mycelium that may form a sclerotium that after overwintering can produce an ascoma or b) crossing two compatible primary mycelia that after plasmogamy form a heterokaryotic secondary mycelium that may produce a sclerotium that finally forms the ascoma. Sclerotia in vitro derived from polysporic cultures have been observed in *Morchella* spp. associated with *Austrocedrus chilensis* in Argentina

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PLATE 12. 1–9. *Morchella elata* (LPS 35912). 1. Ascomata: a, in vertical section. 2. Sketch of a cross section of the pileus: h, hymenium, sh, subhymenium, t, medullary excipulum, A–B, rib. 3. Detail of B in FIG. 2. 4. Sketch of a longitudinal section of the stipe: ce, cortical layer, t, trama, ci, internal layer. 5. Detail of the D in FIG. 2. 6. Detail of B in FIG. 4. 7. Paraphyses. 8. Ascus. 9. Ascospores.





(unpublished results). Spawn and a kit for outdoors cultivation of *Morchella* spp. are now commercially available.

DISTRIBUTION IN ARGENTINA: Five species are recorded in Argentina: *M. esculenta*, *M. intermedia* Boud., *M. elata* Fr., *M. patagonica* Speg., *M. semilibera* DC. from CO, N, RN, TF. There are several unidentified collections preserved in LPS and BCRU.

ILLUSTRATION: Pl. 12, 1–9. *Morchella elata*.

LITERATURE: Boudier 1897; Buscot 1992, 1993; Buscot & Kottke 1990; Dahlstrom et al. 2000; Dissing 2000; Domínguez de Toledo 1987; Eckblad 1968; Hennebert & Bellemère 1979; Jung et al. 1993; Gamundí 1975; Gamundí & Horak 2003; Jacquetant 1984; Landvik et al. 1997; O'Donnell et al. 1997; Ower 1982; Paden 1972; Parguey-Leduc et al. 1998; Rifai 1968; Volk & Leonard 1990.

### *Nothojafnea* Rifai (Pyronemataceae)

ASCOMATA apothecial, small- to medium-sized, superficial, sessile, gregarious, cup shaped; disc deeply concave, reddish brown to dark brown; external surface brown to reddish brown paler than the disc, felty; hairs short, slender, pauciseptate, straight or curved, hyaline or subhyaline, sometimes with a brownish sap. ECTAL EXCIPULUM a textura angularis of isodiametric or polygonal light brown cells disposed at right angle to the surface, some superficial cells clavate, thick walled, containing brownish sap. MEDULLARY EXCIPULUM of compact textura intricata, composed of hyaline, slender hyphae running horizontally. ASCI subcylindrical, rather thick walled, 8-spored, J–. PARAPHYSES subclavate, simple, containing brownish pigment at the apex, pluriseptate. ASCOSPORES 1-seriate, hyaline, when young multiguttulate, ellipsoidal, ornamented with small warts weakly stained with lactic blue.

TYPE SPECIES: *Nothojafnea cryptotricha* Rifai, Verh. Kon. Ned. Akad. Wetensch., Afd. Natuurk. 2de Reeks, 57(3): 94. 1968.

HABITAT: on soil.

ANAMORPH: unknown.

NOTES: *Nothojafnea* is close to *Jafnea*. The latter genus differs in having brown-walled hairs and fusoid to fusiform-ellipsoidal ascospores. It is also distinct from *Aleurina* in ascospores and hairs. (See description of *Aleurina*.)

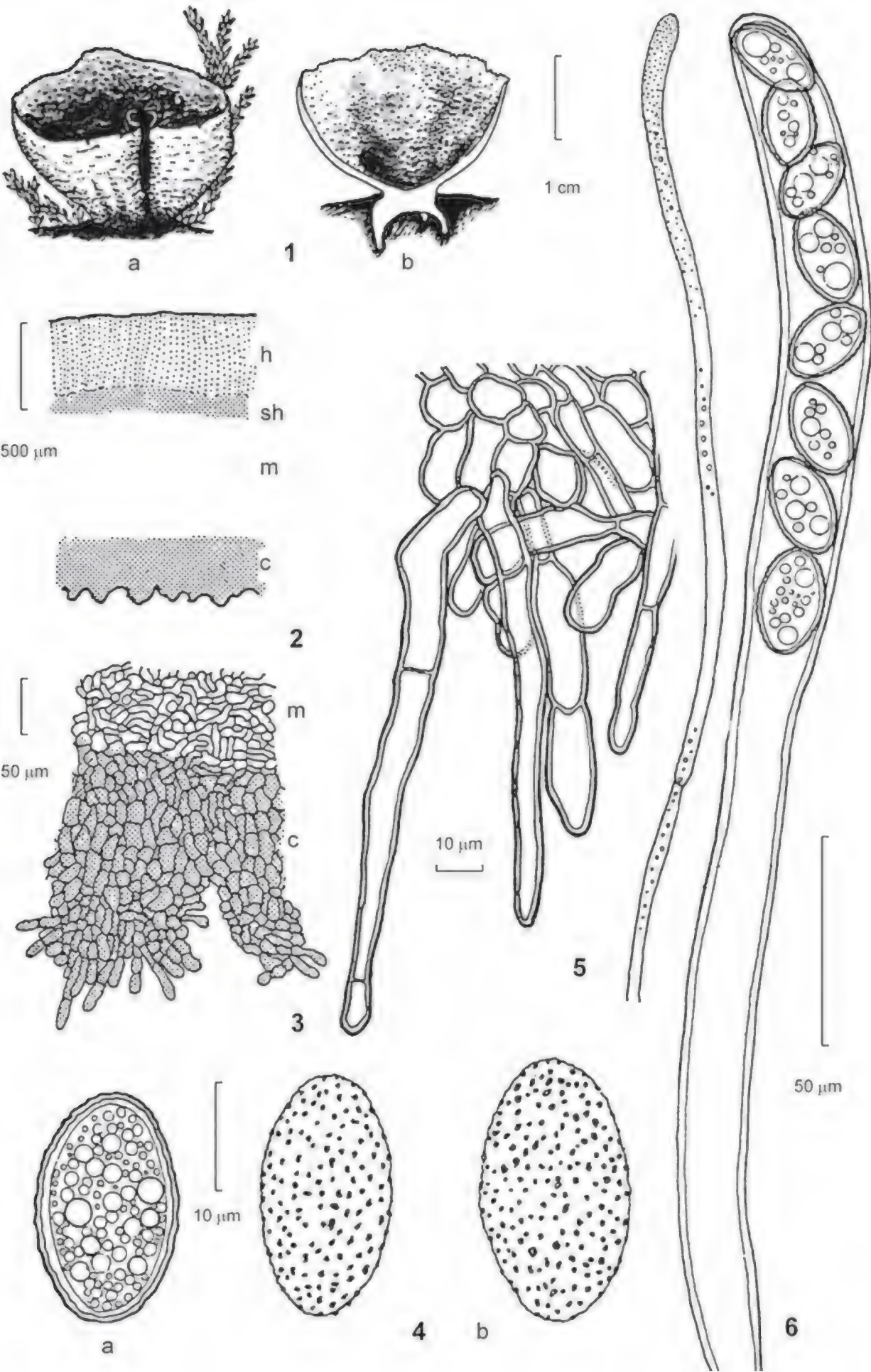
DISTRIBUTION IN ARGENTINA: Only one species is recorded: *N. thaxteri* (Cash) Gamundí from CH, N and RN.

ILLUSTRATION: Pl. 13, 1–6. *Nothojafnea thaxteri*.

LITERATURE: Eckblad 1968; Gamundí 1972a, 1999; Gamundí et al. 2004; Korf 1960, 1973a; Rifai 1968; Zhuang & Korf 1986.

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PLATE 13. 1–6. *Nothojafnea thaxteri* (BAC 5838). 1. Ascomata: a, side view, b, vertical section. 2. Sketch of a vertical section of the ascoma: h, hymenium, sh, subhymenium, m, medullary excipulum, c, ectal excipulum. 3. Detail of the excipulum: m, c, as in FIG. 2. 4. Ascospores: a, optical section, b, surface view. 5. Hairs of the ectal excipulum. 6. Ascus and paraphyses.





*Phillipsia* Berk., nom. cons. (*Sarcoscyphaceae*)

ASCOMATA apothecial, small to large, cup shaped, sometimes asymmetrical, superficial, sessile, subsessile to stipitate, scattered to gregarious, usually bright coloured; disc shallow or deeply concave, smooth or umbilicate, in several shades of orange, pink, reddish, yellow, purplish or brownish violet; exterior furfuraceous to tomentose, paler than the disc, consisting of simple, hyphal, flexuous, superficial, hyaline hairs. ECTAL EXCIPULUM, thin, a textura porrecta to intricata of hyaline hyphae, running more or less parallel to the surface of the receptacle. MEDULLARY EXCIPULUM well developed, of loose textura intricata. ASCI cylindrical to subcylindrical, suboperculate, thick walled, gradually attenuated towards the base, 8-spored, J-. PARAPHYSES pluriseptate, filiform, straight, hyaline, simple or branched, sometimes anastomosing with each other, containing carotenoids (major pigment phillipsiaxanthine). ASCOSPORES 1-seriate, multinucleate, containing 1–2 guttules, hyaline to pale yellowish, ellipsoidal with acute ends to subapiculate, inaequilateral, with longitudinal ridges that occasionally anastomose or wrinkled, and ornamentation arising from the primary wall, cyanophobic.

TYPE SPECIES: *Phillipsia domingensis* (Berk.) Berk., J. Linn. Soc., Bot. 18: 388. 1881.

HABITAT: on fallen angiosperm branches or wood.

ANAMORPH: *Molliardiomyces* Paden. Germinating ascospores produce conidiophores with conidiogenous cells with sympodial or percurrent proliferation, conidia holoblastic, subglobose, hyaline.

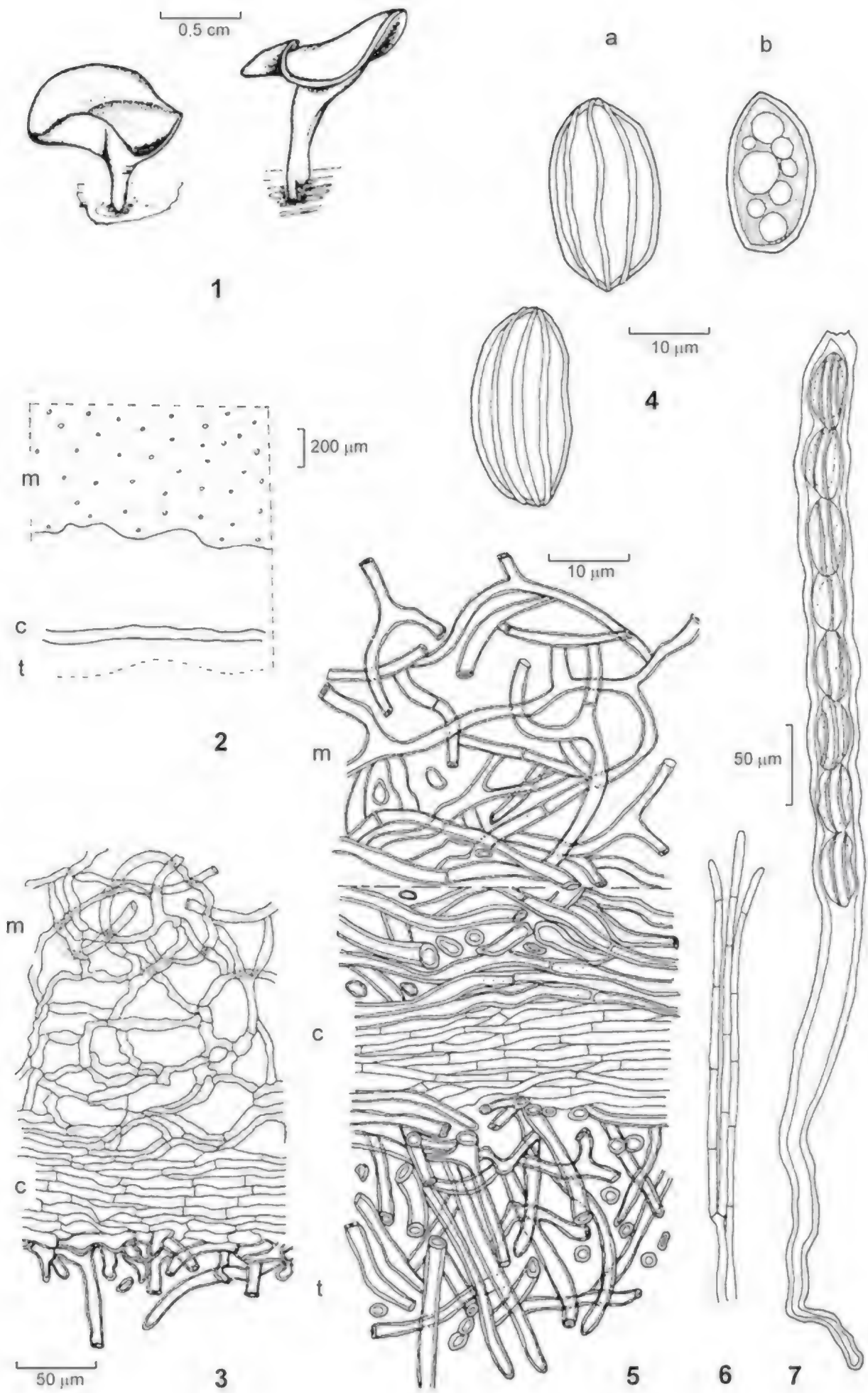
NOTES: *Phillipsia* shows a great variability in disc colour, which in the same species can vary, for example, from deep pink to whitish. Some species contain a particular carotenoid pigment, phillipsiaxanthin. The genus is close to *Cookeina* in sharing similar ascospore ornamentation, suboperculate ascus apex, and the same major pigment but differing in other characters (see NOTES under *Cookeina*).

*Nanoscypha* Denison, also with a *Molliardiomyces* anamorph, differs in excipular structure and 4-spored asci. *Sarcoscypha* (Fr.) Boud. differs in its equilateral, subcylindrical, and always smooth ascospores. The type of ascospore germination is similar to *Sarcoscypha*.

ITS-based molecular studies of *Phillipsia* imply four main lineages that are supported by ascospore morphology: 1) the *P. domingensis* complex, which includes ascospores ornamented with separate and few longitudinal ridges;

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PLATE 14. 1–7. *Phillipsia domingensis* (BAFC 30278). 1. Ascomata. 2. Sketch of a vertical section of the ascoma in the basal zone: m, medullary excipulum, c, ectal excipulum, t, tomentum. 3. Detail of a section of the ascoma at the lateral zone: m and c, as in FIG. 2. 4. Ascospores: a, surface view; b, optical section. 5. Detail of a vertical section of the ascoma at the basal zone: m, c, and t, as in fig 2. 6. Paraphyses. 7. Ascus.



2) *P. olivacea*, with smooth or wrinkled ascospores; 3) *P. crispata*, with fine, profuse longitudinal, parallel ridges; and 4) *P. carnicolor* Le Gal with broad, irregular, longitudinal ridges sometimes anastomosing. It is suggested that colour of the ascomata should be used with caution as a taxonomic character. The genus is pantropical, reaching subtropical areas.

DISTRIBUTION IN ARGENTINA: four species have been recorded: *P. domingensis*, *P. hartmannii* (W. Phillips) Rifai, *P. crispata* (Berk. & M.A. Curtis) Le Gal and *P. olivacea* Rick (the last cited as *P. rugospora* Paden) from M, T.

ILLUSTRATION: Pl. 14, 1–7. *Phillipsia domingensis*.

LITERATURE: Arpin 1969; Boedijn 1933; Cabello 1988; Denison 1969, 1972; Hansen et al. 1999; Harrington et al. 1999; Kirk et al. 2008; Le Gal 1953; Li & Kimbrough 1996b; Paden 1974, 1977, 1984, 1986; Romero & Gamundí 1986; Zhuang & Wang 1998.

### *Plectania* Fuckel (*Sarcosomataceae*)

ASCOMATA apothecial, medium-sized to large, cup shaped, superficial, sessile to substipitate, scattered to gregarious, of tough gelatinous consistency; disc deeply concave, dark brown to black, smooth, gelatinous, drying cracked; external surface felty to tomentose, dark or a little paler than the disc, venose near the base. ECTAL EXCIPULUM thin, a textura globulosa to angularis of isodiametric, brown-walled cells ending in flexuous, brown, thick-walled, densely intertwined hairs. MEDULLARY EXCIPULUM of a loose textura intricata, well developed, composed of hyaline, branched, septate hyphae embedded in a gelatinous matrix. ASCI cylindrical, thick-walled, contracted below forming an appendiculate base, with a central operculum, 8-spored, J–. PARAPHYSES filiform, pluriseptate, sometimes profusely branched near the apex and anastomosing, forming a delicate net, hyaline or containing a diffuse pigment. ASCOSPORES 1-seriate, multinucleate, hyaline to pale yellowish, containing many guttules, ellipsoidal to asymmetrically fusoid or subballantoid, smooth or covered by non-cyanophilic transverse ridges on the convex side that occasionally anastomose.

TYPE SPECIES: *Plectania melastoma* (Sowerby) Fuckel, Jahrb. Nassauischen Vereins Naturk. 23–24: 324. 1870.

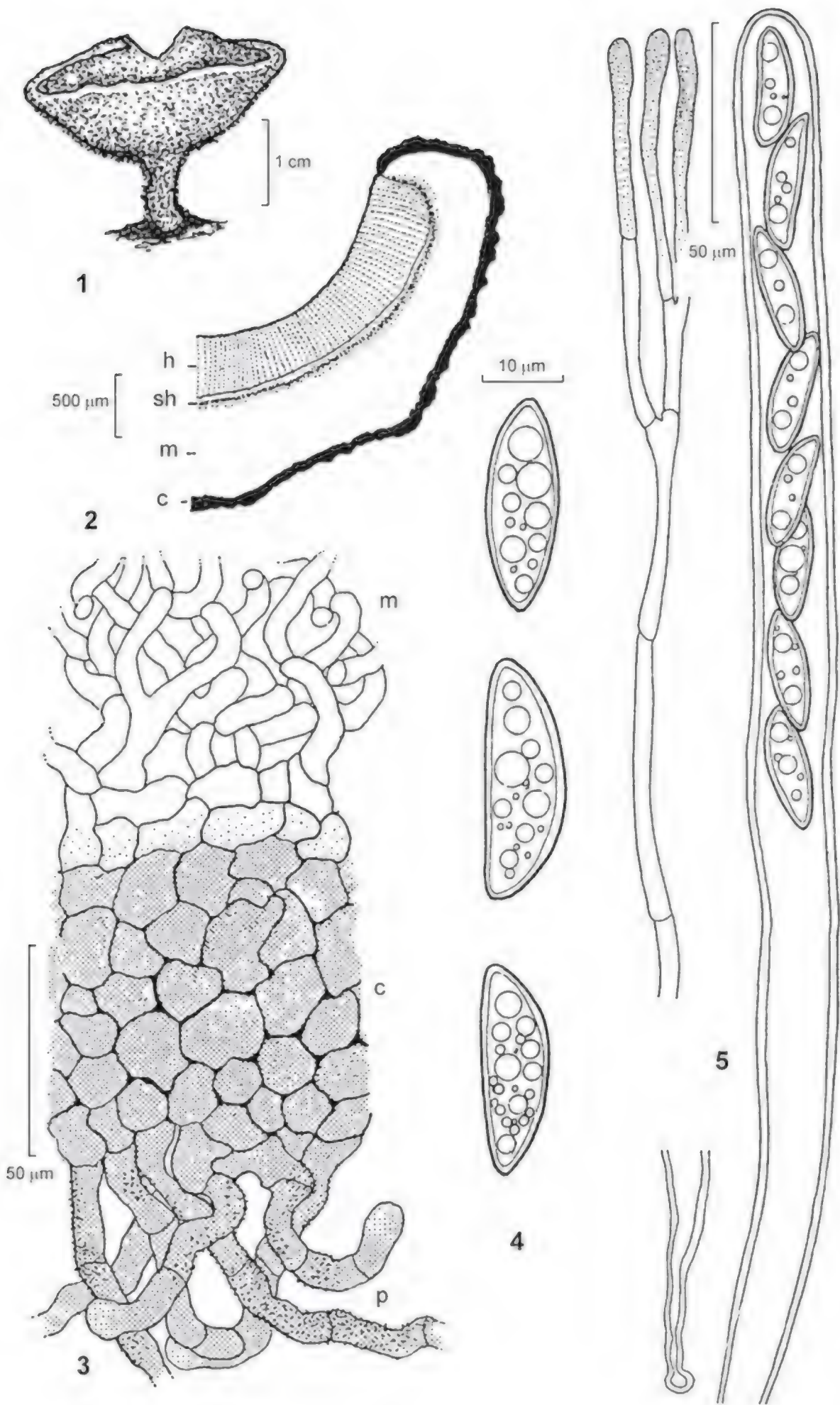
HABITAT: on twigs, plant debris, decaying logs, sometimes covered with mosses, in coniferous and deciduous forests.

ANAMORPH: *Conoplea* Pers. Conidiomata synnematos, pulvinate, sometimes with a stromatic base or groups of conidiophores mononematous scattered on the

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PLATE 15. 1–5. *Plectania chilensis* (Lazo Pu-28). 1. Ascoma. 2. Sketch of a vertical section of the ascoma: h, hymenium, sh, subhymenium, m, medullary excipulum, c, ectal excipulum. 3. Detail of the excipulum, m and c, as in FIG. 2, p, hairs. 4. Ascospores. 5. Ascus base and upper portion and paraphyses.





substratum; conidiophores geniculate, arborescent, brownish; conidiogenous cells sympodioblastic; conidia holoblastic, unicellular, globose to ellipsoidal, brownish, smooth or verrucose, with a slit or pore.

NOTES: According to modern authors, *Plectania* is related to *Urnula* Fr., which also has black, large ascomata but lacks the gelatinous medullary excipulum. Ultrastructural studies (TEM) comparing ascus walls of the *Plectania* and *Urnula* type species show that they have a similar structure. *Pseudoplectania* Fuckel, which is perhaps another close genus, has globose ascospores, with different ontogeny of the walls, as demonstrated by ultrastructural studies (TEM). The mature ascospore wall in *Plectania* is composed of primary wall, episporium, and secondary wall, while *Pseudoplectania* lacks the secondary wall. *Sarcosoma* Casp. differs in its highly gelatinous, turbinate ascomata. *Galiella* Nannf. & Korf is distinguished by ascospore walls with cyanophylic ornamentation. SSU rDNA and 18S rRNA sequence-based analyses support all genera mentioned above in the *Sarcosomataceae*, a monophyletic or paraphyletic family different from the *Sarcoscyphaceae*, as previously suggested from ultrastructure (TEM) of the ascus-wall layers.

DISTRIBUTION IN ARGENTINA: two species are recorded: *P. chilensis* (Mont.) Gamundí and *P. rhytidia* (Berk.) Nannf. & Korf from: BA, CH, M, N, RN.

ILLUSTRATION: Pl. 15, 1–5. *Plectania chilensis*.

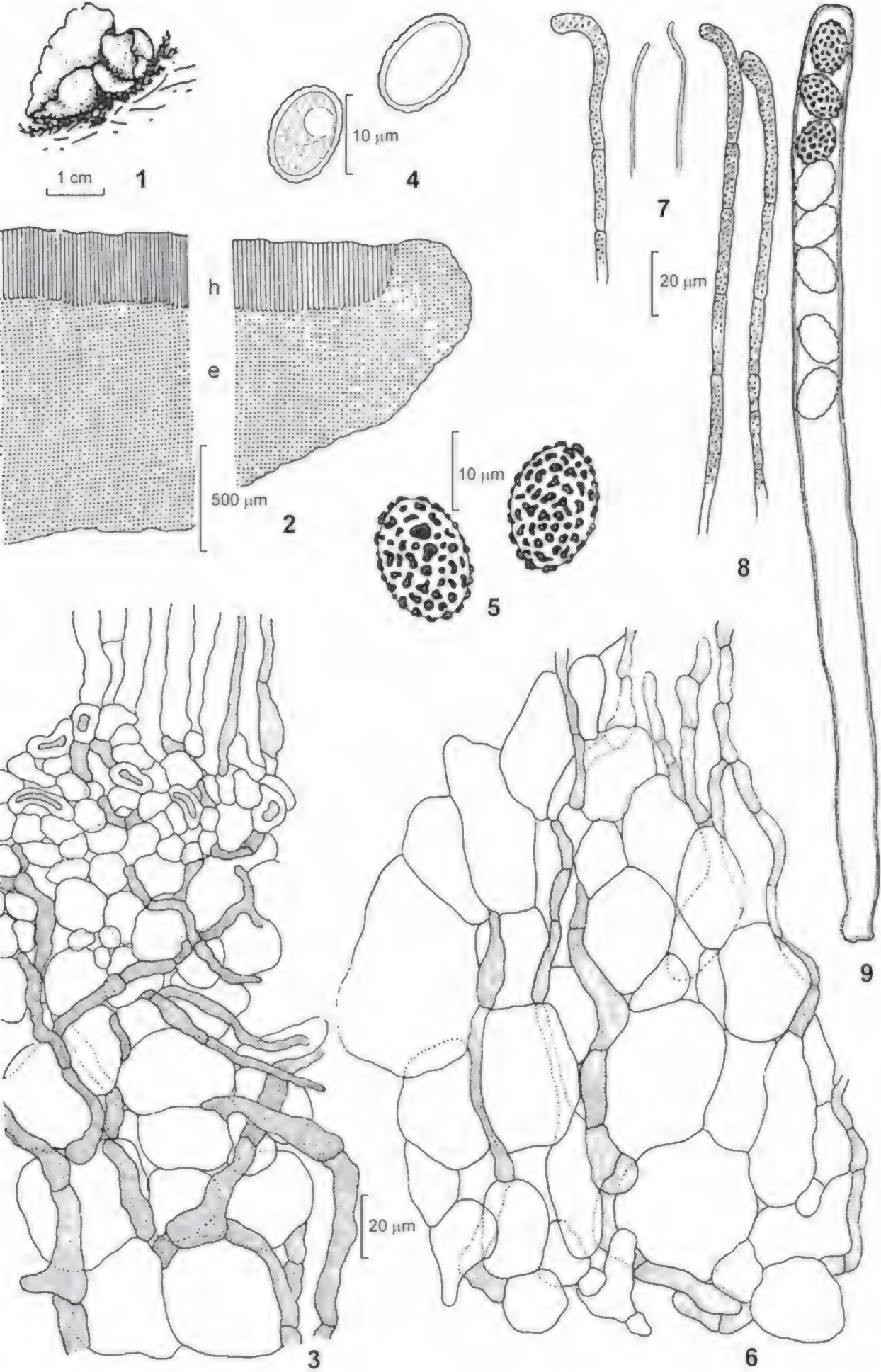
LITERATURE: Bellemère et al. 1990; Benkert 2005; Cabello 1988; Dissing 2000; Donadini 1985; Eckblad 1968; Gamundí 1971; Gamundí & Gaiotti 1998; Gamundí et al. 2004; Harrington et al. 1999; Hughes 1960; Korf 1957, 1973a; Le Gal 1953; Landvik et al. 1997; Li & Kimbrough 1995; Liu & Zhuang 2006; Paden 1972; Rifai 1968; Romero & Gamundí 1986; Sutton & Hennebert 1994.

### ***Rhodopeziza* Hohmeyer & J. Moravec (*Pezizaceae*)**

ASCOMATA apothecial, medium-sized to large, superficial, sessile to subsessile, cupuliform to cochleate; margin pruinose; disc smooth, concave sometimes undulate, orange reddish (*miniatus*); margin conspicuous, pruinose; external surface smooth. ECTAL EXCIPULUM of textura angularis to textura globulosa, composed of cells larger than the medullary cells. MEDULLARY EXCIPULUM a textura globulosa to angularis of cells occasionally intermixed with hyphae. ASCI cylindrical, operculate, 8-spored, the whole wall turning blue with iodine (J+). PARAPHYSES simple, pluriseptate, subclavate and bent towards the apex, containing granules of a carotenoid pigment, that turns green with iodine. ASCOSPORES, 1-seriate, with one evanescent guttule, hyaline to pale yellowish,

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PLATE 16. 1–9. *Rhodopeziza tuberculata* (LPS 37095). 1. Ascomata. 2. Sketch of a vertical section of the ascoma: h, hymenium, e, excipulum. 3. Detail of the excipulum in the internal zone. 4. Immature ascospores. 5. Mature ascospores. 6. Detail of the excipulum in the external zone. 7. Dehiscent ascus. 8. Paraphyses. 9. Ascus.





broadly ellipsoidal, tuberculate, with tubercles isolated, conical to truncate, conspicuously cyanophilic.

TYPE SPECIES: *Rhodopeziza tuberculata* (Gamundí) J. Moravec & Hohmeyer, Czech Mycol. 47(4): 261. 1995 ["1994"].

HABITAT: on soil, among liverworts.

ANAMORPH: unknown.

NOTES: *Rhodopeziza* is very similar to *Aleuria* (*Pyronemataceae*), both macroscopically and microscopically, sharing a brightly coloured hymenium (due to a carotenoid pigment in the paraphyses), subhymenium, and excipular structure. Main differences are the tuberculate (instead of reticulate) ascospores and a weak J+ reaction of the entire ascus wall (compared to J– in *Aleuria*). This character led Hohmeyer and Moravec to create the monotypic genus *Rhodopeziza*. On the other side, if we emphasize the character of carotenoid pigment plus the diffuse iodine reaction of the ascus wall, the closest genus would be *Iodophanus* Korf (*Pezizaceae*). Moravec placed *Rhodopeziza* in the *Pezizales*. Eriksson and Hawksworth, based on the presence of iodine positive asci, decided to incorporate the genus into the *Pezizaceae* while referring *Aleuria* to the *Pyronemataceae*. A J+ ascus wall is currently accepted as a phylogenetically more important character than the pigmentation of the hymenium. It would be desirable to collect the type species again to confirm the iodine positive ascus wall as a character that defines the taxonomic position of the genus.

DISTRIBUTION IN ARGENTINA: *R. tuberculata* was only found in TF cited as *Aleuria tuberculata* Gamundí. It has not been reported elsewhere in the world.

ILLUSTRATION: Pl. 16, 1–9. *Rhodopeziza tuberculata*.

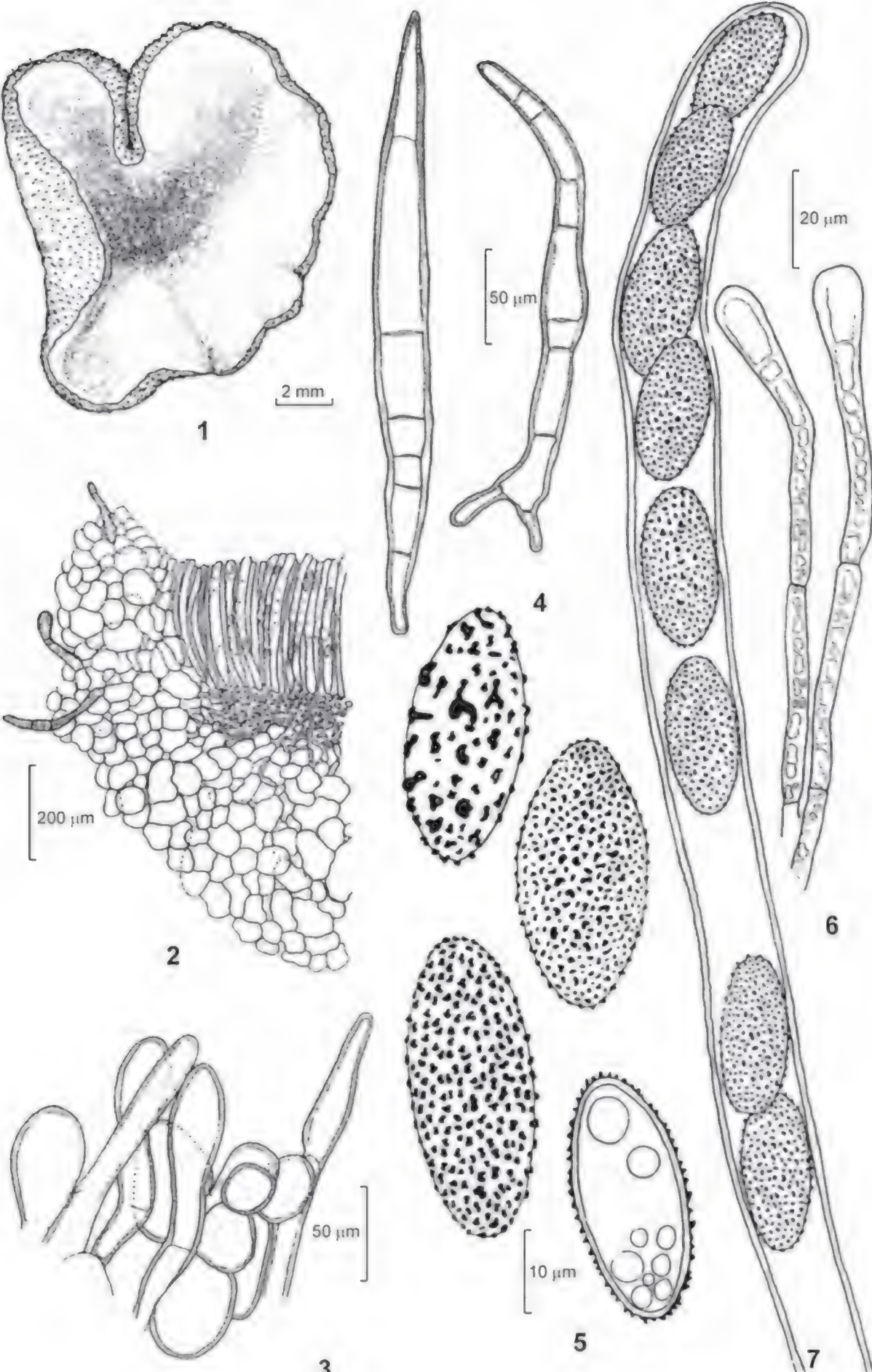
LITERATURE: Eriksson & Hawksworth 1995; Gamundí 1975; Gamundí et al. 2004; Hansen et al. 2001; Häffner 1993; Moravec 1994b.

### *Scutellinia* (Cooke) Lambotte, nom. cons. (*Pyronemataceae*)

ASCOMATA apothecial, small- to medium-sized, superficial, sessile, saucer-shaped, gregarious, usually bright coloured; disc smooth to undulate, from orange, red, to reddish-brown, exceptionally white; margin and external surface hairy, ochraceous to brownish; hairs setose, simple, brown to brownish black, multiseptate, with thick lateral walls, thin septa and forked bases arising deeply from the excipulum, up to 3000 µm long, the marginal hairs longer than the lateral; superficial hairs shorter, brownish, simple, rarely bifurcate at the base. ECTAL EXCIPULUM of textura angularis to textura globulosa. MEDULLARY EXCIPULUM of textura intricata, with hyphae densely arranged horizontally.

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PLATE 17. 1–7. *Scutellinia doelloi* (LPS 35716). 1. Ascoma. 2. Vertical section of the ascoma. 3. Detail of the margin. 4. Setose hairs. 5. Ascospores. 6. Paraphyses. 7. Ascus: upper portion.



ASCI cylindrical, operculate, usually 8-spored, less commonly 2–4 spored, J–. PARAPHYSES multiseptate, straight, club-shaped to pear-shaped at the apex, usually containing granules of carotenoid pigments—major pigment  $\gamma$ -carotene—that turn green in iodine and blue in sulphuric acid in fresh material. ASCOSPORES 1-seriate, uni- to multinucleate, uni- to multiguttulate, globose, ellipsoidal to subfusoidal, hyaline to pale yellowish, ornamented with warts, spines, ridges or a reticulum intensely dying with lactic-blue (cyanophilic), rarely smooth.

TYPE SPECIES: *Scutellinia scutellata* (L.) Lambotte, Mém. Soc. Roy. Sci., Liège, sér. 2, 1: 199. 1887.

HABITAT: on soil, wood and plant debris, in wet places, sometimes associated with mosses and liverworts.

ANAMORPH: unknown.

NOTES: *Scutellinia* is taken in the sense of modern authors to replace *Lachnea* (Fr.) Gillet (typified by an inoperculate discomycete) and *Ciliaria* Quél. (an illegitimate name). The name *Patella* F.H. Wigg. was rejected after Korf & Schumacher's (1986) proposed to designate *Scutellinia* a nomen conservandum. *Scutellinia* is related to *Cheilymenia*, which has also rooting hairs (which can be either brown or hyaline in that genus). Recent nLSU rDNA sequence analyses suggest affinity between these genera, which TEM studies on septal pores also support. (See NOTES under *Cheilymenia*.)

*Scutellinia* differs from *Anthracobia*, which has blunt, non-rooting hairs and biguttulate ascospores. Cultures in PDA may produce mycelium with brown, monilioid chlamydospore chains. Germinating ascospores may give rise to microconidia. According to substrata, they have been classified in three ecological groups: humus saprotrophs, xylosaprotrophs, and forest saprotrophs. A worldwide monograph was provided by Schumacher who used cladistic analysis and proposed an infrageneric classification with two subgenera, *Scutellinia* and *Legalia*, both represented in Argentina. Phylogenetic relationships derived from partial SSU and LSU rDNA sequence data suggest the main core of *Scutellinia* spp. is closely related to *Cheilymenia fimicola*, but the remaining *Cheilymenia* spp. resolve quite distantly and form a group with *Trichophaea*–*Anthracobia*. The carotenoid pigment of the disc is characteristic of *Scutellinia* spp. except in *S. nivea* T. Schumach., which has a pale hymenium. The last character and rooted hairs are shared with *Paratrachophaea* Trigaux, but in that genus the ascospores are smooth or slightly punctuate and eguttulate.

DISTRIBUTION IN ARGENTINA: Species recorded from Patagonia (N, RN, TF): *S. badioberbis* (Cooke) Kuntze, *S. bifurcata* Gamundí, *S. colensoi* Masee ex Le Gal, *S. doelloi* (Speg.) Le Gal, *S. hirta* (Schumach.) Cooke, *S. hirtella* (Rehm) Kuntze, *S. kerguelensis* (Berk.) Kuntze, *S. nigrohirtula* (Svrček) Le Gal, *S. nivalis* (Boud.) Le Gal,



*S. patagonica* (Rehm) Gamundí, *S. setosa* (Nees) Kuntze, *S. torrentis* (Rehm) T. Schumach. (= *S. marginata* Gamundí), *S. scutellata*, *S. trechispora* (Berk. & Broome) Lambotte, *S. umbrata* f. *antarctica* (Rehm) Gamundí, *S. umbrorum* (Fr.) Lambotte. Species recorded from Central and N Argentina: *S. balansae* (Speg.) Gamundí, *S. cubensis* (Berk.) M.A. Curtis, *S. jungneri* (Henn.) Clem. [cited as *S. lurida* (Henn. & E. Nyman) Le Gal] and *S. olivascens* (Cooke) Kuntze (= *S. lusatae* (Cooke) Kuntze) from BA, J, ME, MI.

ILLUSTRATION: Pl. 17, 1–7. *Scutellinia doelloi*.

LITERATURE: Arpin 1969; Denison 1961; Gamundí 1956, 1960, 1964, 1975; Gamundí et al. 2004; Kaushal et al. 1983; Kimbrough & Curry 1986; Korf & Schumacher 1986; Kullman 1982; Le Gal 1966, 1969, 1974; Liu & Zhang 2006; Perry et al. 2007; Pfister 1988; Rifai 1968; Romero & Gamundí 1986; Schumacher 1988, 1990; Svrček 1971; Trigaux 1985; Vooren et al. 2005; Waraich 1977; Wang 1998; Yao & Spooner 1995a; Zhuang & Wang 1998.

### *Sowerbyella* Nannf. (*Pyronemataceae*)

ASCOMATA apothecial, epigeous, medium-sized to large, cupulate, superficial, stipitate, scattered to gregarious, sometimes conrescent at the stipe, of fleshy consistency; disc bright yellow to yellow-orange; margin involute, entire to undulate; external surface tomentose, paler than the disc; stipe cylindrical, longitudinally venose, sometimes enlarged in the middle portion and hollow, half or totally buried in the substratum. ECTAL EXCIPULUM of textura globulosa to angularis with cells arranged in rows perpendicular to the surface, the outermost ending in hyphose, obtuse, septate, hyaline hairs that form the tomentum. MEDULLARY EXCIPULUM well developed, a textura intricata of hyaline hyphae sometimes with swollen articles. ASCI cylindrical tapering below, 8-spored, J–. PARAPHYSES straight, cylindrical or slightly enlarged at the apex, hyaline, pluriseptate. ASCOSPORES 1-seriate, uninucleate, hyaline, ellipsoidal, containing 2 guttules, smooth, verrucose, spiny or reticulate, the reticulum being complete or incomplete and derived from the perispore, cyanophilic.

TYPE SPECIES: *Sowerbyella radiculata* (Sowerby) Nannf., Svensk Bot. Tidskr. 32: 119. 1938.

HABITAT: on damp soil, rotten twigs, and leaves, in woodlands among mosses.

ANAMORPH: unknown.

NOTES: *Sowerbyella* was originally described as having verrucose ascospores and included only two species. It differs from other yellow or orange taxa in paraphyses that do not turn green with iodine and in stipitate ascomata. (See *Aleuria*.) They share the brightly coloured disc, fleshy consistency, and type of ascospore ornamentation. *Otidea* (Pers.) Bonord., which is somewhat related, is distinguished by ascomata that are usually ear-shaped and glabrous and smooth ascospores. A revision of the type species of *Sowerbyella* using SEM

for studying the ascospore ornamentation revealed that the marking forms a complete or incomplete reticulum that can vary throughout the same collection. Molecular studies support this viewpoint and show that *Sowerbyella* forms an isolated clade with an unresolved position in the family *Pyronemataceae*. If this new concept of *Sowerbyella* is accepted, the genus is undoubtedly closely related to *Aleuria*.

DISTRIBUTION IN ARGENTINA: Only one species is recorded from the Andean-Patagonian forest, *S. rhenana* (Fuckel) J. Moravec, from CH, N, RN.

ILLUSTRATION: Pl. 18, 1–8. *Sowerbyella rhenana*.

LITERATURE: Benkert 2005; Eckblad 1968; Gamundí 1960, 1964; Gamundí & Horak 2003; Gamundí et al. 2004; Korf 1972; Moravec 1985, 1988, 1994b; Nannfeldt 1938; Perry et al. 2007; Yao & Spooner 2006; Zhuang 2009.

### *Tricharina* Eckblad emend. Chin S. Yang & Korf (*Pyronemataceae*)

ASCOMATA apothecial, small- to medium-sized, deeply cupulate to discoid, of fleshy consistency, gregarious, sessile, superficial and broadly sessile to partially sunken in the substrate; disc, smooth, white, gray, yellow, orange to brown; margin conspicuous, hairy, with fascicles of hairs arising from the outermost cells of the excipulum, simple, straight or flexuous, pluriseptate, acute or obtuse at the apex with basal cells usually inflated, hyaline, subhyaline or brown-walled. ECTAL EXCIPULUM a textura angularis to globulosa of hyaline or subhyaline cells or the outermost layers with brown-walled cells. MEDULLARY EXCIPULUM of textura intricata, hyaline. ASCI cylindrical, usually 8-spored, J–. PARAPHYSES simple, slightly enlarged at the apex. ASCOSPORES usually 1-seriate, uninucleate, eguttulate, but sometimes with polar granules, hyaline, immature ascospores with the cytoplasm staining blue in cotton blue, at maturity very refractive, yellowish with a cyanophylic sheath discernible with cotton blue, ellipsoidal to subfusoidal, smooth or ornamented with fine warts, sometimes arranged in longitudinal stripes.

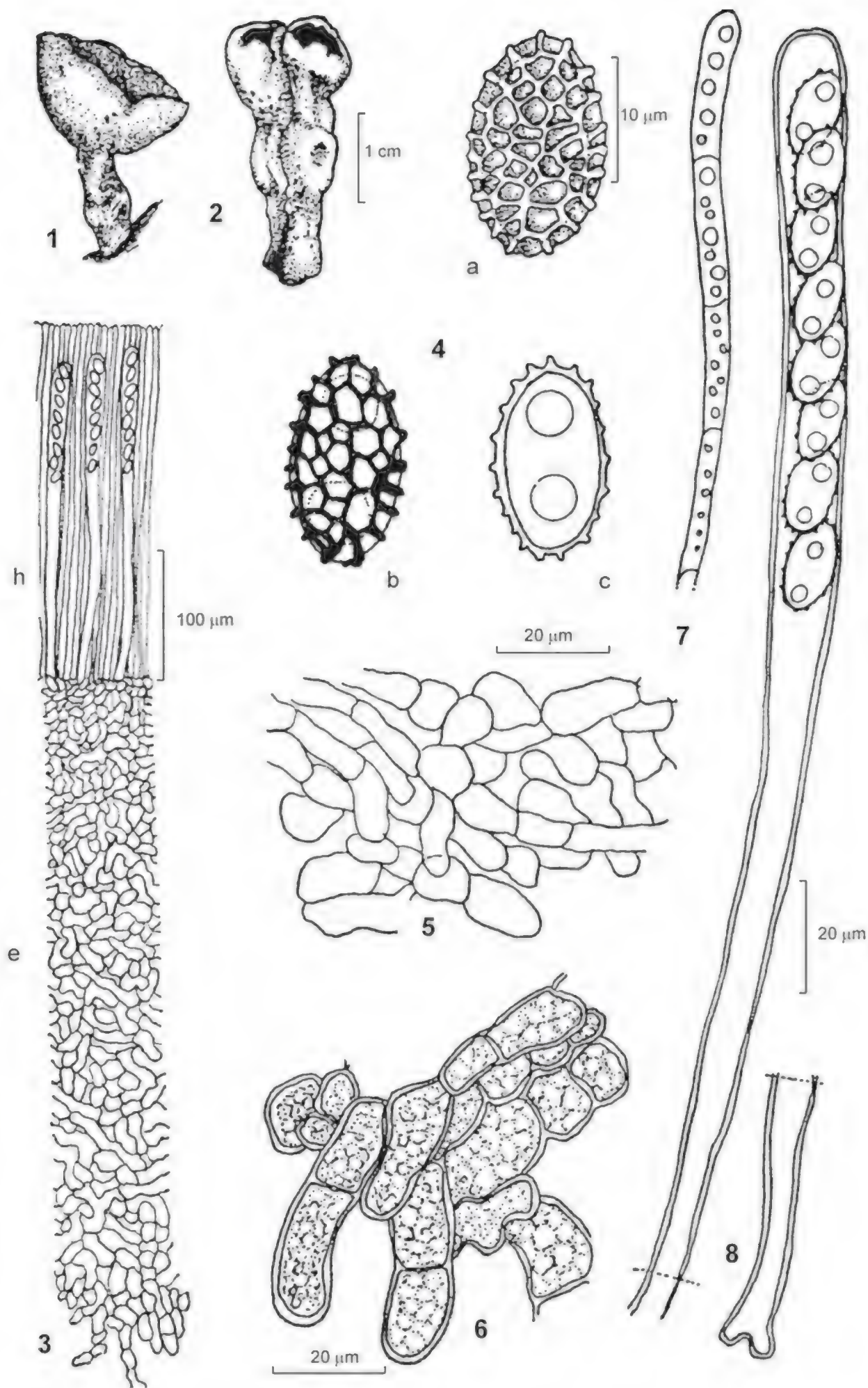
TYPE SPECIES: *Tricharina gilva* (Boud. ex Cooke) Eckblad, Nytt Mag. Bot. 15: 60. 1968.

HABITAT: on burnt soil, decayed wood and plant debris.

ANAMORPH: *Ascorhizoctonia* Chin S. Yang & Korf, a *Rhizoctonia*-like anamorph. Mycelium superficial or embedded in the agar-media, hyaline to brownish. Forms aggregates of monilioid, branched cell-chains; cells doliiform containing oil globules.

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PLATE 18. 1–8. *Sowerbyella rhenana* (BAFC 20579). 1. Mature ascoma. 2. Concescent young ascomata. 3. Vertical section of the ascoma: h, hymenium, e, excipulum. 4. Ascospores: a, surface view unstained, b, idem. stained with lactic blue, c, optical section. 5. Detail of the excipulum. 6. Excipular furfurations. 7. Paraphyses. 8. Ascus: upper and lower portions.





NOTES: *Tricharina* is similar to *Trichophaea* (see below) and is also related to *Wilcoxina* Chin S. Yang & Korf, which differs in hairs that cover the entire receptacle down to the base and a narrower, uninflated basal cell. Its anamorph is the chlamydosporic *Complexipes* C. Walker emend. Chin S. Yang & Korf, which forms ectomycorrhizae.

*Trichophaeopsis*, a segregate of *Trichophaea* that is also similar but characterized by bifurcate hairs, is distinguished by an ectal excipulum formed by horizontally elongated, thick-walled brown cells and ascospores lacking oil globules but with de Bary bubbles.

The relationship of *Tricharina* with *Geopora* is supported by molecular studies using partial nLSU rDNA and SSU rDN sequences from various species of both genera that differ morphologically in ascospore guttulation and hair morphology (see description of *Geopora*). The figures given to illustrate the genus *Tricharina* were published as *Trichophaea fimbriata* (Quél.) Gamundí (1966) after examination of the type specimen in Cooke's Herbarium (K). In the revision of *Tricharina* by Yang & Korf (1985), *T. fimbriata* is considered a synonym of *T. gilva* for nomenclatural reasons, a view that I accept. However, the Argentine collection has ascospores that agree in form and size [ $15\text{--}16.6\text{--}18.3 \times 8.3\text{--}10 \mu\text{m}$ ] with the type specimen of *Lachnea fimbriata* Quél. Brummelen (1983) stated that *T. gilva* is very variable in ascospore size and length/breadth ratio.

DISTRIBUTION IN ARGENTINA: two species are recorded: *T. gilva* (= *Trichophaea fimbriata* (Boud. ex Cooke) Gamundí and *T. striispora* (Rifai) Chin S. Yang & Korf, from BA and RN.

ILLUSTRATION: Pl. 19, 1–7. *Tricharina gilva*.

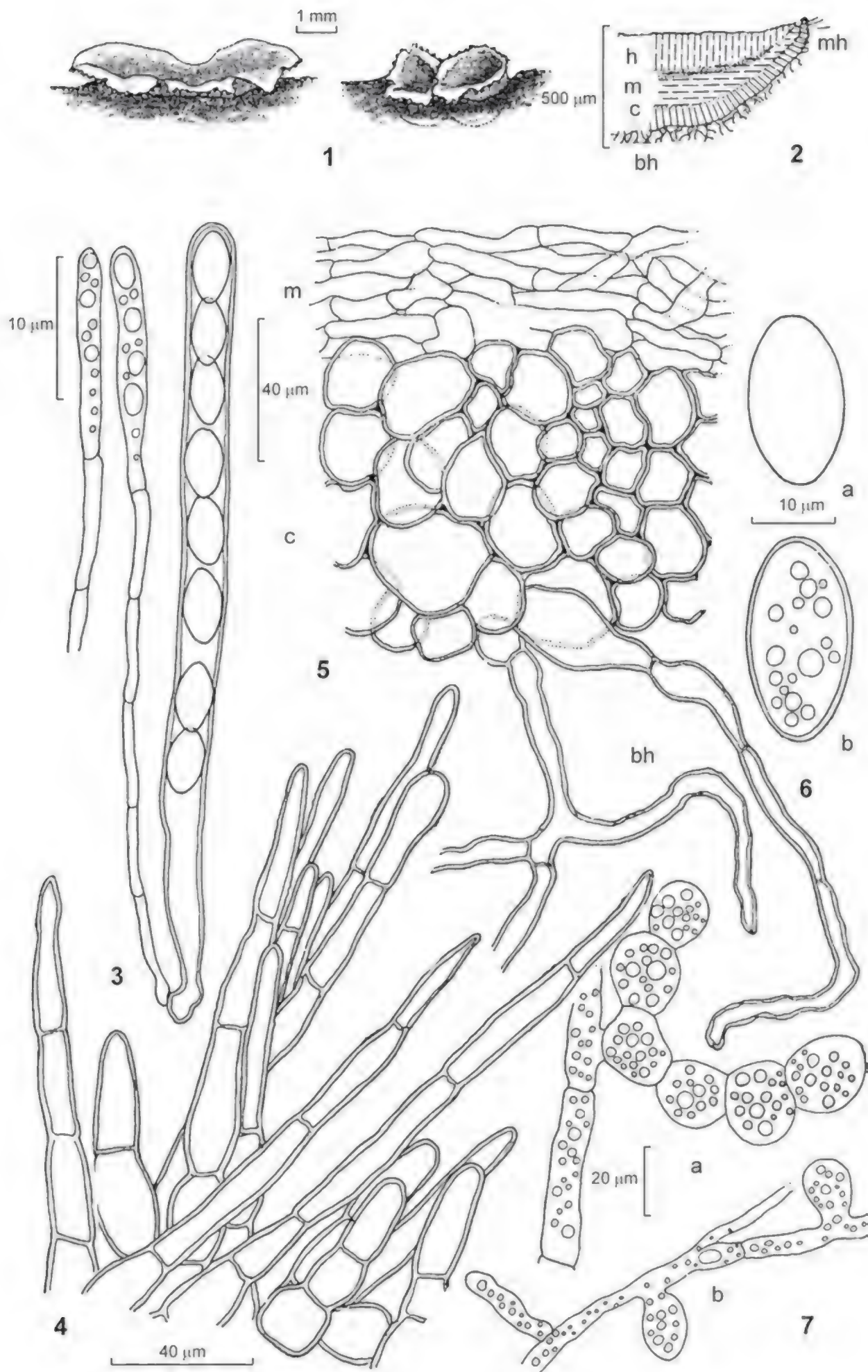
LITERATURE: Barrera & Romero 2001; Brummelen 1983; Dennis 1978; Dissing 2000; Eckblad 1968; Egger 1996; Gamundí 1966, 1975; Gamundí & Lorenzo 2001; Gamundí et al. 2004; Korf 1985; Korf & Erb 1972; Liu & Zhuang 2006; Perry et al. 2007; Svrček & Kubička 1961; Wu & Kimbrough 1996; Yang & Korf 1985a,b; Yang & Kristiansen 1989.

### *Trichophaea* Boud. (Pyronemataceae)

ASCOMATA apothecial, small- to medium-sized, sessile, discoid to pateliform, gregarious, of fleshy consistency; disc smooth, plane to concave, whitish, pale bluish, grayish to pale ochraceous grayish; margin conspicuous, hairy, covered with scattered, long, superficial hairs, isolated or in fascicles, acute and rigid, pluriseptate, thin-walled, hyaline, yellowish or brown, simple sometimes with

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PLATE 19. 1–7. *Tricharina gilva* (BAFC 22002). 1. Ascomata. 2. Sketch of a vertical section of the ascoma: h, hymenium, m, medullary excipulum, c, ectal excipulum, bh, basal hairs, mh, marginal hairs. 3. Ascus and paraphyses. 4. Marginal hairs. 5. Vertical section of the ascoma: m, c and bh as in FIG. 2. 6. Ascospores: a, surface view, b, optical section. 7. Immerse mycelia in a 4-week culture at room temperature: a, filter paper medium, chlamydospore-like cells, b, APG medium, young mycelium.



a bulbous base or attenuated at the base; external surface concolorous with the disc or brownish. ECTAL EXCIPULUM of textura angularis to globulosa, composed of isodiametric cells with hyaline or brown walls, arranged in rows perpendicular to the external surface, the most superficial sometimes forming patches of brown cells from where the hairs arise. MEDULLARY EXCIPULUM of textura intricata, hyaline. ASCI cylindrical, 8-spored, J–. PARAPHYSES simple, clavate at the apex, hyaline, septate. ASCOSPORES 1-seriate, uninucleate, hyaline, subglobose, ellipsoidal to fusoid, smooth or ornamented with small to large warts, guttulate, sometimes with de Bary bubble.

TYPE SPECIES: *Trichophaea woolhopeia* (Cooke & W. Phillips) Arnould, Bull. Soc. Mycol. France 9: 112. 1893.

HABITAT: on clayish or burnt soil, on plant debris and mushroom beds.

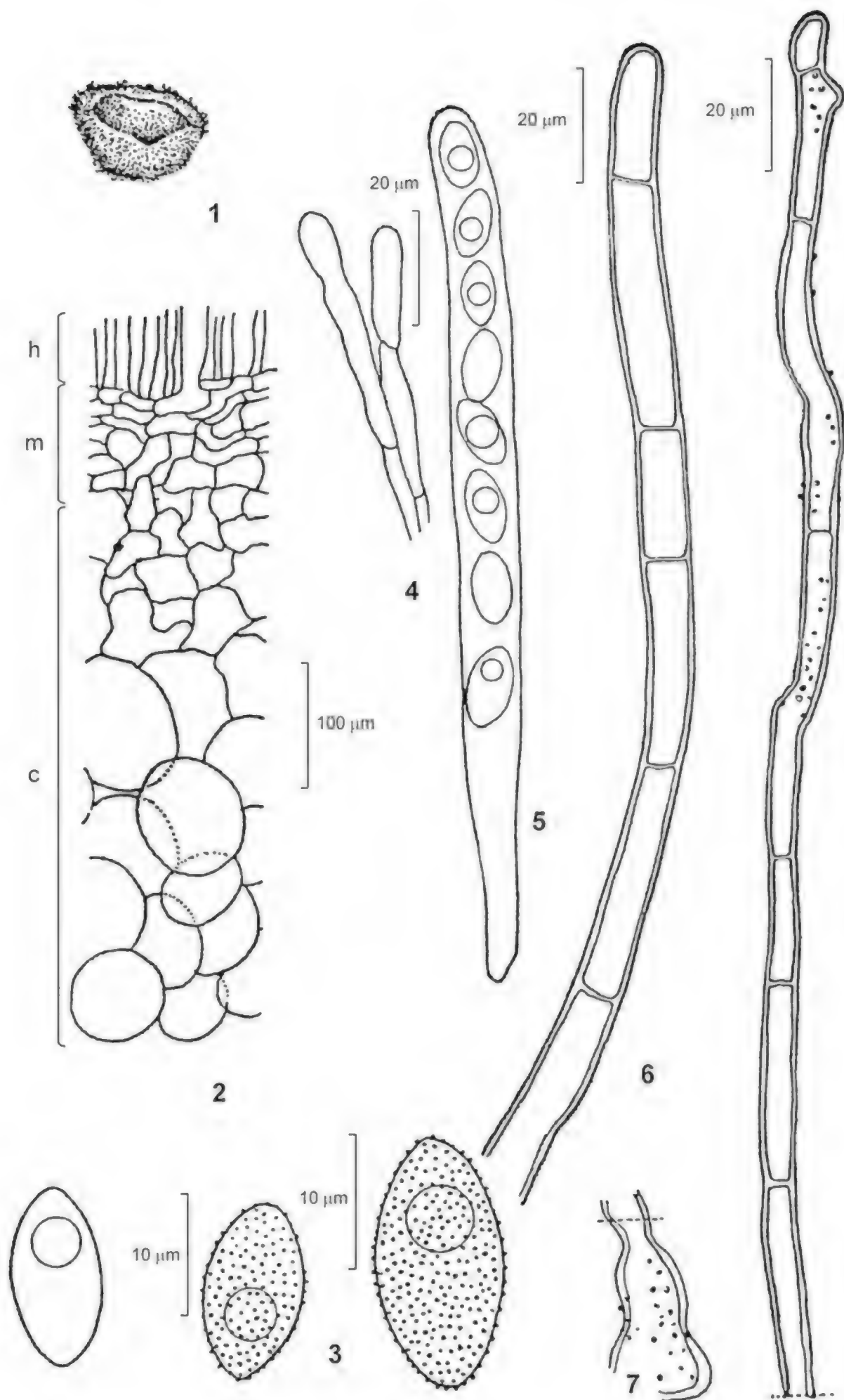
ANAMORPH: *Dichobotrys* Hennebert. Conidiophores mononematose, hyaline, erect, dichotomously furcate at about half height, with primary and secondary branches; conidiogenous cell sympodioblastic; conidia holoblastic, hyaline, unicellular, subglobose to napiform, smooth.

NOTES: *Trichophaea* is related to other hairy *Pyronemataceae* such as *Anthracobia* but differs in its long, pointed hairs (see *Anthracobia* above). Some species of both genera may colonize burnt places but can be distinguished at first sight by the colour of the disc. Also related is *Sphaerosporella*, which has globose, uniguttulate ascospores. Ultrastructure examination of the ascospore wall led some authors to reunite *Sphaerosporella* and *Trichophaea*. Both genera also have the same anamorph genus, *Dichobotrys*. *Trichophaeopsis* differs in its bifurcate hairs and superficial excipular cells arranged in vertical rows. *Tricharina* also shows affinity with *Trichophaea*, which has an *Ascorhizoctonia* anamorph. *Paratrachophaea* differs in its setiform hairs arising deeply in the excipulum and eguttulate ascospores. Some species of pyrophilous *Trichophaea* can complete the life cycle in vitro but ascospores may need to be submitted to a heat shock to stimulate germination. Others, non-pyrophilous, can form ectomycorrhiza with *Betula* and *Picea*, a symbiosis confirmed by experimental and molecular studies. Ultrastructural septal structure (TEM) showed that *Trichophaea* has the aleurioid-type of ascospores (see NOTES in *Aleuria*). Results on ascospore ontogeny demonstrated that smooth-spored and pyrophilous species form *Dichobotrys* anamorphs, whereas rough-spored species are non-pyrophilous and do not form anamorphs. Phylograms generated from SSU rDNA data analyses suggest affinity with *Wilcoxina*, which has *Complexipes* anamorphs.

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PLATE 20. 1–6. *Trichophaea gregaria* (LIL, Singer T-2266). 1. Ascoma. 2. Vertical section of the ascoma: h, hymenium, m, medullary excipulum, c, ectal excipulum. 3. Ascospores in optical section and surface view. 4. Paraphyses. 5. Ascus. 6. Marginal hair. 7. Basal hair, base and terminal portion.





Partial nLSU rDNA sequence analyses suggest that *Trichophaea* is non-monophyletic. A clade “7”, with smooth ascospores and *Dichobotrys* anamorph suggest a close connection with *Sphaerosporella*, while clade “8” (diagnosed by ornamented ascospores and without any anamorph) is related to *Wilcoxina*.

DISTRIBUTION IN ARGENTINA: one species was found: *T. gregaria* (Rehm). Boud. from BA.

ILLUSTRATION: Pl. 20, 1-7. *Trichophaea gregaria*.

LITERATURE: Coetzee & Eicker 1994; Dennis 1978, 1981; Dissing 2000; Gamundí 1960; Hennebert 1973; Hennebert & Bellemère 1979; Kanouse 1958; Kimbrough 1994; Korf 1988; Korf & Erb 1972; Landvik et al. 1997; Liu & Zhuang 2006; Maas Geesteranus 1967; Perry et al. 2007; Pfister 1988; Rifai 1968; Tedersoo et al. 2005; Trigaux 1985; Svrček & Kubička 1961; Vooren et al. 2005; Webster et al. 1964; Wu & Kimbrough 1995; Yang & Korf 1985a,b.

### *Trichophaeopsis* Korf & Erb (*Pyronemataceae*)

ASCOMATA apothecial, minute to small, turbinate, gregarious, of fleshy consistency, sessile; disc plane or concave, whitish to dull yellow; margin elevated, undulate, hairy; external surface densely covered by straight, dark brown, acute, thick-walled, pluriseptate setae, some of them bifurcate usually with two unequal branches, the longest pointing upwards, lower part of the ascoma covered with flexuous hyaline to brownish, thin-walled, simple hairs, with a bulbous base. Setae and hairs are of superficial origin. ECTAL EXCIPULUM of textura prismatica, one- or two-layered, composed of thick-walled, brown cells, in surface view horizontally elongated. MEDULLARY EXCIPULUM well developed, of a compact textura intricata, hyaline. ASCI cylindrical, 4–8-spored, J–. PARAPHYSES simple, filiform, hyaline. ASCOSPORES usually 1-seriate, uninucleate, eguttulate, smooth or punctuate, hyaline or pale yellowish and very refractive, sometimes with a de Bary bubble.

TYPE SPECIES: *Trichophaeopsis bicuspis* (Boud.) Korf & Erb, Phytologia 24(1): 18. 1972.

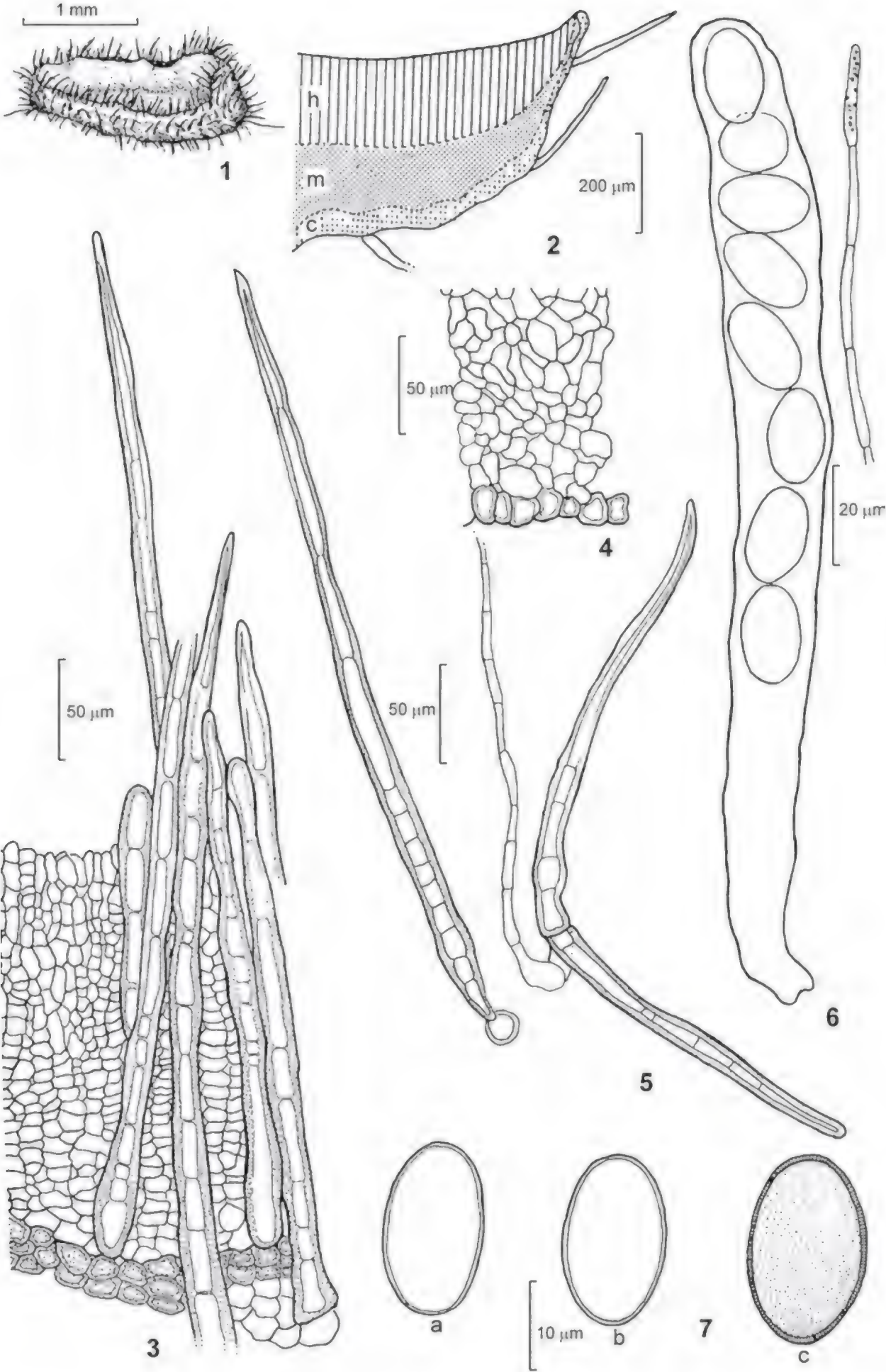
HABITAT: on dung, soil or plant debris.

ANAMORPH: unknown.

NOTES: *Trichophaeopsis* is characterized by its very thin, dark ectal excipulum and bifurcate setae that separate it from *Trichophaea* with simple hairs and different type of ectal excipulum (compare descriptions). The genus, which includes two species (and one subspecies), it is said by its authors to occupy an isolated position among the operculate discomycetes. It was recently suggested

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PLATE 21. 1–7. *Trichophaeopsis bicuspis* subsp. *eguttulispora* (LPS 36891). 1. Ascoma. 2. Sketch of a vertical section of the ascoma: h, hymenium, m, medullary excipulum, c, ectal excipulum. 3. Margin and ectal excipulum in surface view. 4. Detail of the ectal excipulum. 5. Setiform and hyphoid hairs. 6. Ascus and paraphyses. 7. Ascospores: a, b unstained, c, iodine stained.





that its closest relative is *Rhizoblepharia* Rifai. This relationship seems remote, as hairs in this genus are rooting, the ascospores are fusoid and covered with transverse, cyanophobic ridges recalling those of some *Sarcoscyphaceae*. *Wilcoxina* is different in excipular structure and hairs. *Paratrachophaea* differs basically in its simple setae that arise in the medullary excipulum. Molecular studies using parsimony and Bayesian analysis of partial sequences of SSU and LSU rDNA suggests a relationship of *Trichophaeopsis* with *Wilcoxina* and a group of *Trichophaea* spp. The synonymy of *Trichophaea gilva* (Boud. ex Cooke) Gamundí, a homotypic synonym of *Tricharina gilva*, with *Trichophaea eguttulispora* Gamundí that appeared in Gamundí et al. (2004: 141) is erroneous; the correct name of the latter taxon is *Trichophaeopsis bicuspis* subsp. *eguttulispora* (Gamundí) Korf.

ILLUSTRATION: Pl. 21, 1–7. *Trichophaeopsis bicuspis* subsp. *eguttulispora*.

DISTRIBUTION IN ARGENTINA: only *T. bicuspis* subsp. *eguttulispora* from TF.

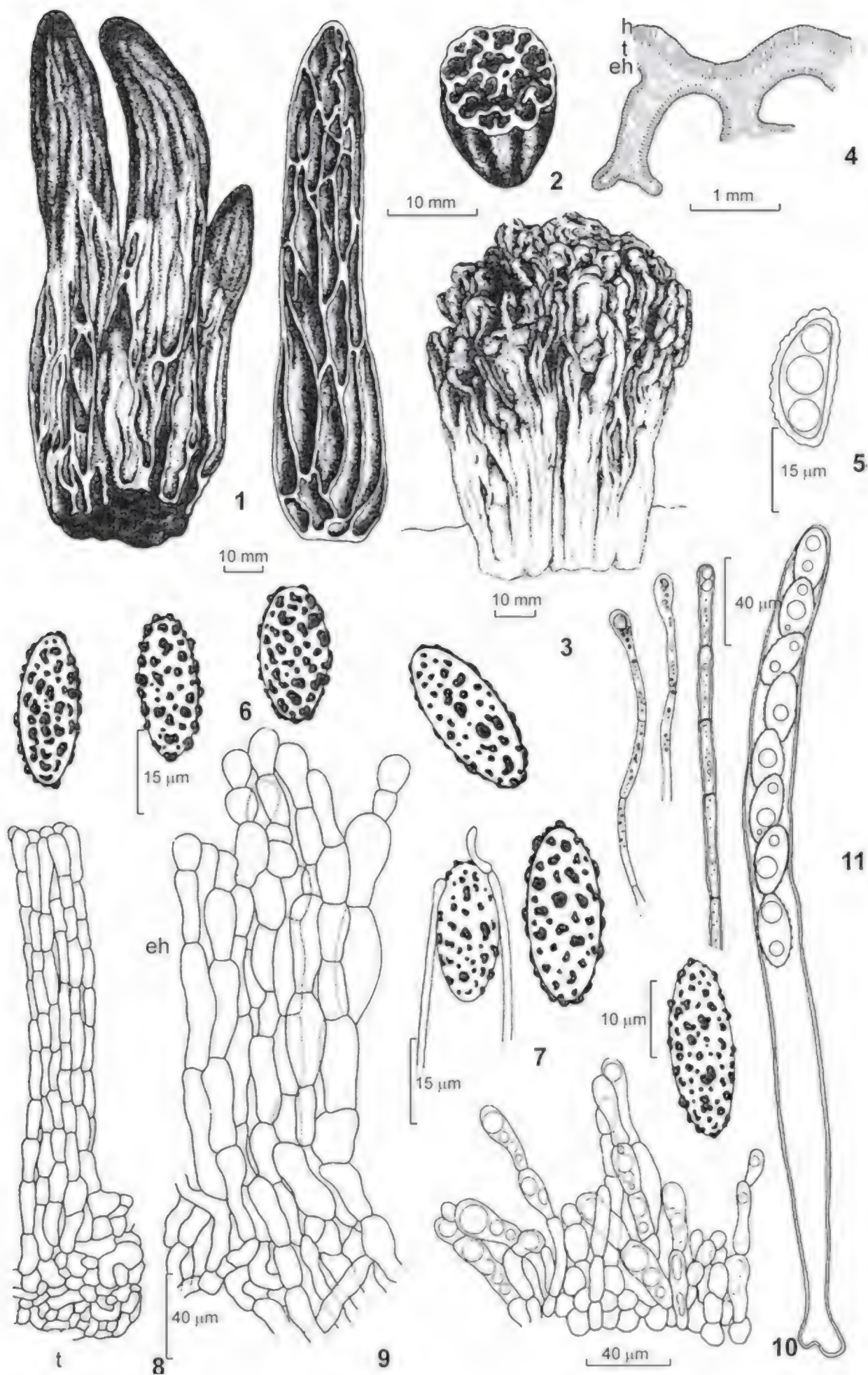
LITERATURE: Barrera & Romero 2001; Dissing 2000; Dissing & Paulsen 1976; Gamundí 1975; Gamundí et al. 2004; Häffner & Krieglsteiner 1991; Hansen & Pfister 2006; Korf 1977; Korf & Erb 1972; Landvik et al. 1997; Liu & Zhuang 2006; Perry & Pfister 2008; Perry et al. 2007; Pfister 1988; Trigaux 1985; Vooren et al. 2005; Yang & Korf 1985a;b; Yang & Kristiansen 1989.

### *Underwoodia* Peck (*Helvellaceae*)

ASCOMATA pileate, cylindrical or clavate, straight or slightly curved, large, up to 25 cm long, superficial, stipitate with the pileus completely adnate to the stipe, gregarious to cespitose, sometimes concrescent, of fleshy consistency drying leathery, internally entirely hollow or alveolate; hymenium brown, grayish-brown or black, covering the upper part of the ascoma, smooth or sulcate; stipe cylindrical, somewhat bulbous at the base, smooth or sulcate with longitudinal ribs that may anastomose, internally hollow or lacunose and externally minutely furfuraceous, paler than the hymenium or whitish. PILEUS in transverse section showing: a) hymenium as the outermost layer, followed by b) a trama of compact textura intricata and c) a palisade-like inner layer of textura prismatica. STIPE 3-layered, composed of: a) External palisade-like layer composed of septate hyphae disposed in rows perpendicular to the surface, the outermost ending freely to form the furfurations; b) Medium layer (trama) of textura intricata of hyaline, septate hyphae; c) Inner layer palisade-like of hyaline hyphae, similar

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PLATE 22. 1–11. *Underwoodia fuegiana* (BAFC 20001). 1. Ascomata, one in vertical section. 2. Cross section of the ascoma at the pileus level. 3. Several concrescent ascomata. 4. Sketch of a cross section of the pileus: h, hymenium, t, trama, eh, palisade-like layer. 5. Ascospore, optical section. 6. Ascospores, surface view. 7. Dehiscent ascus. 8. Detail of the palisade-like layer of the pileus: eh, t, as in FIG. 4. 9. Detail of the palisade-like layer of the stipe. 10. Detail of stipe furfuration. 11. Ascus and paraphyses.



to the inner layer of the pileus. ASCI cylindrical to subcylindrical, 8-spored, pleurorhynchous, J-. PARAPHYSES straight or curved and slightly enlarged at the apex, sometimes forked near the base, containing pigmented granules or diffused pigment. ASCOSPORES 1–2 seriate, 4-nucleate, containing 1–3 guttules, hyaline to subhyaline, ellipsoidal to subfusoidal, coarsely verrucose or papulose, warts rounded of unequal size, cyanophilic.

TYPE SPECIES: *Underwoodia columnaris* Peck, Ann. Rep. N.Y. St. Mus. 43: 32. 1890.

HABITAT: on soil in the forest or in disturbed prairies nearby forests, sometimes among mosses and ferns, occasionally on wood.

ANAMORPH: unknown.

NOTES: *Underwoodia* is the legitimate name for *Geomorium* Speg. It shares with *Helvella* the character of 4-nucleate ascospores but distinct by its adnate pileus and ascospore rougher ornamentation. Formerly several authors suggested the synonymy with *Helvella* but others considered it a separate genus. Recent studies confirm the identity of *Underwoodia* as a genus (See NOTES under *Helvella*). Moreover, phylogenetic relationships derived from molecular studies suggest that *Underwoodia* diverges from *Helvella* and from the hypogeous genera *Barssia* and *Balsamia*, all included in the family *Helvellaceae*. Species of North America are recorded as poisonous but no data on this respect have been recorded for the Argentinian collections. No type of mycorrhiza has been confirmed for this genus, but our personal field observation on *U. singeri* Gamundí & E. Horak shows rhizomorphs arising from the base of the stipe which are in contact with the root system of vascular plants. *U. fuegiana* occasionally shows a sparassoid form derived from confluent ascomata when growing in grazing land.

DISTRIBUTION IN ARGENTINA: two species are recorded in the Andean-Patagonian forests: *U. fuegiana* (Speg.) Gamundí and *U. singeri*. from: N, RN, TF.

ILLUSTRATION: Pl. 22, 1–11. *Underwoodia fuegiana*.

LITERATURE: Ammirati et al. 1985; Abbott & Currah 1997; Dissing 1966; Eckblad 1968; Gamundí 1957b, 1975; Gamundí & Horak 1979, 2003; Korf 1973a; Landvik et al. 1999; O'Donnell et al. 1997; Rifai 1968.

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**Notes on *Trametes* from the Brazilian Amazonia**

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**Abstract** — *Trametes supermodesta* is reported as new to Brazil and the collection represents the second from South America. *Trametes ochroflava* and *T. pavonia* represent first records for the Brazilian Amazonia. A description of *T. supermodesta* and a key to the accepted species of *Trametes* reported for the Brazilian Amazonia are provided.

**Key words** — Polyporaceae, diversity

**Introduction**

*Trametes* is a cosmopolitan genus proposed by Fries and comprises about 48–50 species so far ([www.indexfungorum.org](http://www.indexfungorum.org); Kirk et al. 2008). The species of *Trametes* cause white rot of dead hardwood and (rarely) conifers. The genus is characterized by its sessile to effused–reflexed, light-colored basidiomata, poroid hymenial surface with round, angular to irregular pores, trimitic hyphal system, presence or absence of cystidia, and ellipsoid to allantoid, hyaline, smooth basidiospores that do not react in Melzer's reagent (Ryvarden & Gilbertson 1993).

Despite the high biodiversity of the Brazilian Amazonia, the knowledge about *Trametes* is still scarce, with only nine species reported: *T. cotonea*, *T. cubensis*, *T. lactinea*, *T. marianna* (Pers.) Ryvarden 1973, *T. maxima*, *T. membranacea*, *T. modesta*, *T. pubescens* (Schumach.) Pilát 1939, and *T. villosa* (Gomes-Silva & Gibertoni 2009). *Trametes supermodesta* was first described from Venezuela (Ryvarden & Iturriaga 2003), and we provide a description of the species based on collections from the Brazilian Amazonia, a key of the species of the genus in the area, and comments on the species recently collected or deposited in INPA.



## Material and methods

The Amazonia covers an area of 4,196,943 km<sup>2</sup> out of which approximately 50% belongs to Brazil (Capobianco et al. 2001), in the states of Acre, Amapá, Amazonas, Pará, Roraima, Rondônia, half of Mato Grosso (54%), and part of Maranhão (34%) and Tocantins (9%) (IBGE 2003).

Field trips were undertaken four times from 2007 to 2008 in the state of Rondônia and four times from 2006 to 2008 in the state of Pará. In Rondônia, the study areas were located in Estação Ecológica de Cuniã (08°04'S 63°31'W) of the city of Porto Velho, the state capital and Parque Natural Municipal de Porto Velho (08°45'S 63°54'W). Both areas are covered mostly by open ombrophilous forest and transition forest with savanna. In Pará, the Estação Científica Ferreira Penna (1°44'S 51°27'W) includes typical Amazonian ecosystems and its flora is one of the richer in the Amazonian basin (Lisboa 2002). Additionally, three areas in Rondônia were also visited at irregular intervals, and specimens deposited in INPA were also studied.

The basidiomata were analyzed macro- (shape, color, hymenial surface) and micromorphologically (hyphal system, presence/absence and measurements of sterile structures and basidiospores). Microscopical observations were made from slide preparations with 5% KOH, stained with 1% of aqueous phloxine, and Melzer's reagent (Ryvardeen 1991). Color designations follow Watling (1969). The specimens are deposited in the HFSL and in URM.

## Results

Thirteen species of *Trametes* are reported for the Brazilian Amazonia. *Trametes supermodesta*, previously was only from its type locality, is reported for the second time. Although recently described, several earlier collections had already been deposited in INPA. *Trametes ochroflava* and *T. pavonia* are new records for the Brazilian Amazonia and, together with another seven species that are new records for individual Brazilian States in Amazonia, were studied only from collections deposited in INPA, underscoring the importance of herbaria revisions and accessibility of herbaria records.

## Taxonomy

*Trametes supermodesta* Ryvardeen & Iturr., Mycologia 95(6): 1074 (2003).

Basidiomata annual, pileate, semicircular to flabelliform with a contracted base, solitary or gregarious, up to 3.5–5 cm wide and 2.3–3.5 cm high, 0.2 mm thick, slightly flexible. Abhymenial surface glabrous, dull, concentrically zonate, slightly sulcate, cinnamon (10) to buff (52). Margin entire, acute, concolorous with the abhymenial surface. Context homogeneous, fibrous, thin, up to 0.1 mm thick, cinnamon (10) to buff (52), red in KOH. Tubes more or less concolorous with the pore surface, thin, up to 0.1 mm thick. Hymenial surface with angular pores next to the margin and irregular to slightly decurrent in the rest of the hymenial surface, 2–3 per mm, fawn (29) to clay pink (30).

Hyphal system trimitic; generative hyphae hyaline to yellow, clamped, thin-walled, 2–3.5  $\mu\text{m}$ ; skeletal hyphae yellow, thick-walled, 3–5  $\mu\text{m}$ ; binding hypha hyaline to yellow, thick-walled to solid, 2–3.5  $\mu\text{m}$ . Cystidia absent. Basidia not observed. Basidiospores cylindrical, hyaline, thin-walled, smooth, inamyloid, 8–9  $\times$  2.8–3.5  $\mu\text{m}$ .

SUBSTRATE — on deciduous wood.

MATERIAL EXAMINED: BRAZIL. Amazonas: loc. n. det., 10.VII.1971, G.T. Prance et al. 14035-14074 (INPA 32250, INPA 32289); 24.VII.1971, G.T. Prance et al. 14545 (INPA 32761); 16.IX.1980, B. Lowy et al. 185-196 (INPA 100113, INPA 100083); Presidente Figueiredo, 27.II.1985, C. Dick 678 (INPA 185927); Manaus, 17.II.1990, M.A. de Jesus 1454 (INPA 192699, as *T. modesta*); 17.VII.1990, M.A. de Jesus 1449 (INPA 192695, as *T. modesta*); Pará: Oriximinã, 27.VI.1980, V.L.R. Bononi 618 (INPA 103601); 28.VI.1980, V.L.R. Bononi 658 (INPA 103622); 30.VI.1980, V.L.R. Bononi 816 (INPA 103723); Rondônia: loc. n. det., 4.VII.1968, K.P. Dumont et al. 63-65 (INPA 65103, INPA 65105, as *T. scabrosa*); 6.VII.1968, K.P. Dumont et al. 98 (INPA 65136, as *T. scabrosa*); 23.V.1984, R.D. Goos et al. 1631 (INPA 125136); 1.VI.1984, R.D. Goos et al. 1719 (INPA 125221); Porto Velho, Parque Natural Municipal de Porto Velho, VII.2007, A.C. Gomes-Silva 06-60 (URM 79570, URM 79579); Estação Ecológica de Cuniã, II.2007, A.C. Gomes-Silva 236 (URM 79578); Roraima: Alto Alegre, 10.VI.1986, K.F. Rodrigues et al. 885-895 (INPA 143282, INPA 143289); 10.VI.1986, E.S.S. da Silva 410 (INPA 154906, INPA 154940); loc. n. det., 18.VI.1986, B. Lowy et al. 2069 (INPA 145354, as *Daedalea* sp.).

REMARKS: *Trametes supermodesta*, first described from Venezuela by Ryvarden & Iturriaga (2003), is recognized by its large pores and long basidiospores. *Trametes supermodesta* may be mistaken for *T. modesta* due to its similar basidiomata color, but the pores are larger (2–3 per mm) in *T. supermodesta* than in *T. modesta* (6–10 per mm). The Brazilian specimens of *T. supermodesta* differ macroscopically from the original description by the smaller pores (2–3 per mm vs. 3–4 per mm in the original) and thinner basidiomata.

**Key to the species of *Trametes* recorded from the Brazilian Amazonia**

- 1a. Context with black lines ..... 2
- 1b. Context without black lines ..... 4
- 2a. Pores daedaloid, 2–3 per mm, basidiospores 4.5–5.5  $\mu\text{m}$  long ..... *T. maxima*
- 2b. Pores regular or lacerate, 1–5 per mm, basidiospores 5–8.5  $\mu\text{m}$  long ..... 3
- 3a. Pores dentate to lacerate, 1–3 per mm, basidiospores cylindrical to allantoid  
5.5–8.5  $\times$  2.5–3.5  $\mu\text{m}$  ..... *T. villosa*
- 3b. Pores angular to circular, 4–5 per mm, basidiospores cylindrical  
5–6  $\times$  1.5  $\mu\text{m}$  ..... *T. versicolor*
- 4a. Abhymenial surface with reddish cuticle from the base ..... *T. cubensis*
- 4b. Abhymenial surface without reddish cuticle from the base ..... 5
- 5a. Abhymenial surface azonate or slightly zoned ..... 6
- 5b. Abhymenial surface strongly zoned ..... 9

- 6a. Abhymenial surface tomentose to finely pubescent, context not reacting  
in KOH..... *T. pubescens*
- 6b. Abhymenial surface velutine to glabrous, context reacting in KOH .....7
- 7a. Basidiomata white to cream, context dark brown in KOH, basidiospores  
cylindrical-ellipsoid  $4-7.5 \times 2-3 \mu\text{m}$  ..... *T. lactinea*
- 7b. Basidiomata pale pinkish brown, context red in KOH, basidiospores cylindrical 8
- 8a. Pores 6–10 per mm, basidiospores  $4-6 \times 1.5-2 \mu\text{m}$  ..... *T. modesta*
- 8b. Pores 2–3 per mm, basidiospores  $8-9 \times 2.8-3.5 \mu\text{m}$  ..... *T. supermodesta*
- 9a. Basidiomata sessile to effused-reflexed, basidiospores cylindrical .....10
- 9b. Basidiomata sessile, basidiospores cylindrical-ellipsoid to ellipsoid .....12
- 10a. Basidiomata ochraceous to brown, abhymenial surface smooth to tuberculate,  
glabrous, context homogeneous, basidiospores  $4 \mu\text{m}$  long. .... *T. ochroflava*
- 10b. Basidiomata whitish to cream, abhymenial surface finely velutine to tomentose,  
context cottony or fibrous, basidiospores up to  $3.5 \mu\text{m}$  long .....11
- 11a. Context cottony, pores 3–5 per mm, dissepiments entire, basidiospores  
 $7-11 \times 2.5-3.5 \mu\text{m}$  ..... *T. cotonea*
- 11b. Context fibrous, pores 5–6 per mm, dissepiments lacerate to dentate,  
basidiospores  $4.5-6 \times 2-2.5 \mu\text{m}$  ..... *T. membranacea*
- 12a. Abhymenial surface glabrous, pores round, basidiospores cylindrical-ellipsoid,  
 $6-7 \times 2-2.5(-3) \mu\text{m}$  ..... *T. marianna*
- 12a. Abhymenial surface tomentose, pores angular, basidiospores ellipsoid,  
 $5-6 \times 3-4 \mu\text{m}$  ..... *T. pavonia*

***Trametes cotonea*** (Pat. & Har.) Ryvarden, Norw. JI Bot. 19: 236 (1972).

= *Polyporus cotoneus* Pat. & Har., Bull. Soc. mycol. Fr. 9: 208 (1893).

MATERIAL EXAMINED: BRAZIL. Amazonas: Presidente Figueiredo, 3.IV.1984, M.A. de Jesus 390 (INPA 185336, as *T. nivosa*); Rondônia: loc. n. det., 27.X.1979, R. H. Petersen 273 (INPA 110762, as *Polyporus* sp.); Roraima: Caracará, 16.XI.1977, I. de J. Araújo et al. 437-506 (INPA 76964, INPA 77217); loc. n. det., 30.XI.1977, I. de J. Araújo et al. 733 (INPA 78452, as *T. membranacea*).

DESCRIPTION — Ryvarden & Johansen (1980).

DISTRIBUTION — Pantropical (Ryvarden & Johansen 1980, Ryvarden 2000). In Brazil, reported for the states of Acre, Pará (Gomes-Silva & Gibertoni 2009), and now for the states of Amazonas, Rondônia and Roraima.

NOTES — This species can be recognized in the field by the flexible, cream basidiomata. Macroscopically it is similar to *T. membranacea* but differs by shorter basidiospores.

***Trametes cubensis*** (Mont.) Sacc., Syll. Fung. 9: 198 (1891).

= *Polyporus cubensis* Mont., Annls Sci. Nat., Bot., sér. 2, 8: 364 (1837).

MATERIAL EXAMINED: BRAZIL. Acre: loc. n. det., 10.X.1980, B. Lowy et al. 585 (INPA 100437, as *Polyporus* sp.); 26.X.1980, B. Lowy et al. 988 (INPA 100762, as *Polyporus* sp.); Amazonas: loc. n. det., 6.XI.1977, E.M.L. Freire 158 (INPA 70059); 22.V.1978, R. Singer &



I.J. Araújo 11033 (INPA 76881, as *Microporellus* sp.); 1.VIII.1979, A.C. Webber 62 (INPA 84271, as *Fomitopsis* sp.); 16.IX.1980, B. Lowy et al. 170 (INPA 100084, as *Polyporus* sp.); Presidente Figueiredo, 21.IX.1983, M.A. de Jesus 31-32 (INPA 183649, INPA 183650); Fonte Boa, 1.XI.1986, E.S.S. da Silva et al. 923 (INPA 155037); Pará: Itaituba, 29.IX.1977, M. A. Sousa 8-38 (INPA 84083, INPA 84082, as "*Fomitopsis cubensis*"); Rondônia: loc. n. det., 29.VI.1968, K.P. Dumont et al. 12 (INPA 64827, as *T. scabrosa*); Porto Velho, Parque Natural Municipal de Porto Velho, VII.2007, A.C. Gomes-Silva 276 (URM 79554); Roraima: loc. n. det., 24.VII.1974, G.T. Prance et al. 21386-21366 (INPA 112093, as *Polyporus phlebeius*, INPA 45341); Boa Vista, 21.XI.1977, L. de L. J. Aguiar et al. 665 (INPA 78384).

DESCRIPTION — Gilbertson & Ryvarden (1987).

DISTRIBUTION — Neotropical, and subtropical areas of the USA (Gilbertson & Ryvarden 1987). In Brazil, reported for the states of Pará (Gomes-Silva & Gibertoni 2009), Bahia, Paraná, Pernambuco, Rio Grande do Sul, São Paulo and Santa Catarina (Baltazar & Gibertoni 2009). It is a new record for the states of Acre, Amazonas, Rondônia and Roraima.

NOTES — This species can be recognized in the field by the dimidiate basidiomata with a reddish cuticle from the base.

***Trametes lactinea*** (Berk.) Sacc., Syll. Fung. 6: 343 (1888).

= *Polyporus lactineus* Berk., Ann. Mag. nat. Hist. 10: 373 (1842).

MATERIAL EXAMINED: BRAZIL. Acre: loc. n. det., 11.X.1980, B. Lowy et al. 644 (INPA 100484, as *Polyporus* sp.); Amazonas: Manaus, 1.II.1992, M.A. de Jesus 1523 (INPA 192732); 28.IV.1996, K. Vohland 1808 (INPA 216386, as *T. menziesii*); Pará: Melgaço, VIII.2007, T. B. Gibertoni (URM 79949, URM 79950); II.2008, T. B. Gibertoni (URM 79951); Rondônia: Porto Velho, Bairro Arigolândia, VII.2007, A.C. Gomes-Silva 41 (URM 79557); Estação Ecológica de Cuniã, VII.2008, A.C. Gomes-Silva 568-584 (URM 79555, URM 79556); Fazenda Mucum, VII.2007, A.C. Gomes-Silva 106-156-261 (URM 79558, URM 79564, URM 79566); Parque Natural Municipal de Porto Velho, II.2007, A.C. Gomes-Silva 05-04-14 (URM 79561, URM 79562, URM 79569); VII.2007, A.C. Gomes-Silva 62-259 (URM 79563, URM 79565); II/2008, A.C. Gomes-Silva 461-462-450-455 (URM 79559, URM 79560, URM 79567, URM 79568).

DESCRIPTION — Núñez & Ryvarden (2001).

DISTRIBUTION — Pantropical (Núñez & Ryvarden 2001). In Brazil, recorded in the state of Pará (Gomes-Silva & Gibertoni 2009). It is a new record for the states of Acre, Amazonas and Rondônia.

NOTES — The glabrous abhymenial surface and variable brown color of the basidiomata are similar to those of *Lenzites elegans* (Spreng.) Pat., but this species is macroscopically different due to its thicker basidiomata and the lamellate to sinuous hymenial surface.

***Trametes maxima*** (Mont.) A. David & Rajchenb., Mycotaxon 22(2): 315 (1985).

= *Irpex maximus* Mont., Annls Sci. Nat., Bot., sér. 2, 8: 364 (1837).

MATERIAL EXAMINED: BRAZIL. Acre: loc. n. det., 24.IV.1971, G.T. Prance et al. 12411 (INPA 30734); Amazonas: loc. n. det., 4.IV.1978, R.B. Singer & I. de J. Araújo 10930

(INPA 76880, as *Irpex* sp.); Roraima: Caracará, 16.XI.1977, I. de J. Araújo et al. 421 (INPA 76948, as *Coriolus maximus*).

DESCRIPTION — Gilbertson & Ryvarden (1987).

DISTRIBUTION — Neotropical, also known in subtropical areas of the USA (Gilbertson & Ryvarden 1987). In Brazil, recorded in the state of Amapá and Pará (Gomes-Silva & Gibertoni 2009). It is a new record for the states of Acre, Amazonas and Roraima.

NOTES — The hydroid hymenial surface and the context with black zone characterize this species.

***Trametes membranacea*** (Sw.) Kreisel, Monografias, Ciências, Univ. Habana, Ser. 4, 16: 83 (1971).

≡ *Boletus membranaceus* Sw., Fl. Ind. Occid. 3: 1922 (1806).

MATERIAL EXAMINED: BRAZIL. Amazonas: Manaus, 22.I.1978, I. de J. Araújo et al. 976 (INPA 78748, as *Coriolus* sp.); 22.VI.1985, M.A. de Jesus 726 (INPA 185959); 1.VI.1990, M.A. de Jesus 1392 (INPA 192659); Pará: Itaituba, 29.IX.1977, M.A. de Sousa & L.F. Coêlho 55 (INPA 74633, as *Coriolus pinsitus*).

DESCRIPTION — Gilbertson & Ryvarden (1987).

DISTRIBUTION — Neotropical, also known in subtropical areas of the USA and Argentina (Gilbertson & Ryvarden 1987). In Brazil, recorded in the state of Amapá, Pará (Gomes-Silva & Gibertoni 2009), Bahia, Minas Gerais, Paraíba, Paraná, Pernambuco, Rio Grande do Sul, Santa Catarina (Baltazar & Gibertoni 2009) and now found in Amazonas.

NOTES — This species is characterized by the papyraceous, flabelliform, cream to beige basidiomata. It is similar to *T. pavonia*, but differing by the cylindrical basidiospores.

***Trametes modesta*** (Kunze) Ryvarden, Norw. JI Bot. 19: 236 (1972).

≡ *Polyporus modestus* Kunze, in Weigelt, Surinam Exsiccati (1828)

MATERIAL EXAMINED: BRAZIL. Acre: Rio Branco, 24.IX.1980, B. Lowy et al. 247 (INPA 100178, as *Polyporus* sp.); 7.X.1980, B. Lowy et al. 510-511 (INPA 100427, INPA 100478, as *Polyporus* sp.); 9.X.1980, B. Lowy et al. 554 (INPA 100407, as *Polyporus* sp.); 20.X.1980, B. Lowy et al. 819 (INPA 100669, as *Polyporus* sp.); 24.X.1980, B. Lowy et al. 906 (INPA 100777, as *Polyporus* sp.); 1.XI.1980, B. Lowy et al. 1018 (INPA 100842, as *Polyporus* sp.); loc. n. det., 27.IX.1980, B. Lowy et al. 309 (INPA 100279, as *Polyporus* sp.); 28.IX.1980, B. Lowy et al. 332 (INPA 100233); 4.XI.1980, B. Lowy et al. 1102-1094 (INPA 100866, INPA 100928, as *Polyporus* sp.); Amazonas: Aripuanã, 23.IV.1985, K.F. Rodrigues et al. 307 (INPA 128981, as *Polystictus* sp.); Barcelos, 14.II.1984, G.J. Samuels et al. 303 (INPA 129337); 17.II.1984, G.J. Samuels et al. 354 (INPA 129388); 19.II.1984, G.J. Samuels et al. 458 (INPA 129486); 28.II.1984, G.J. Samuels et al. 545 (INPA 129569); 29.II.1984, G.J. Samuels et al. 592 (INPA 129612); Itacoatiara, 14.XI.1966, G.T. Prance et al. 3175 (INPA 18727); 31.XII.1966, G.T. Prance et al. 3628 (INPA 19214); 23.VII.1968, K.P. Dumont et al. 147 (INPA 65183, as *Coriolus* sp.); Manaus, 13.V.1977, M.A. de Sousa 150 (INPA 74656, as *Coriolopsis byrsina*); 10.IX.1977, M.A. de Sousa & I. de J. Araújo 147 (INPA 74654, as *Coriolopsis byrsina*); 1.XI.1977, E.M. de L. Freire 1 (INPA 92688, as *Coriolopsis byrsina*); 28.VI.1978, R. B. Singer & I. de J. Araújo 11266

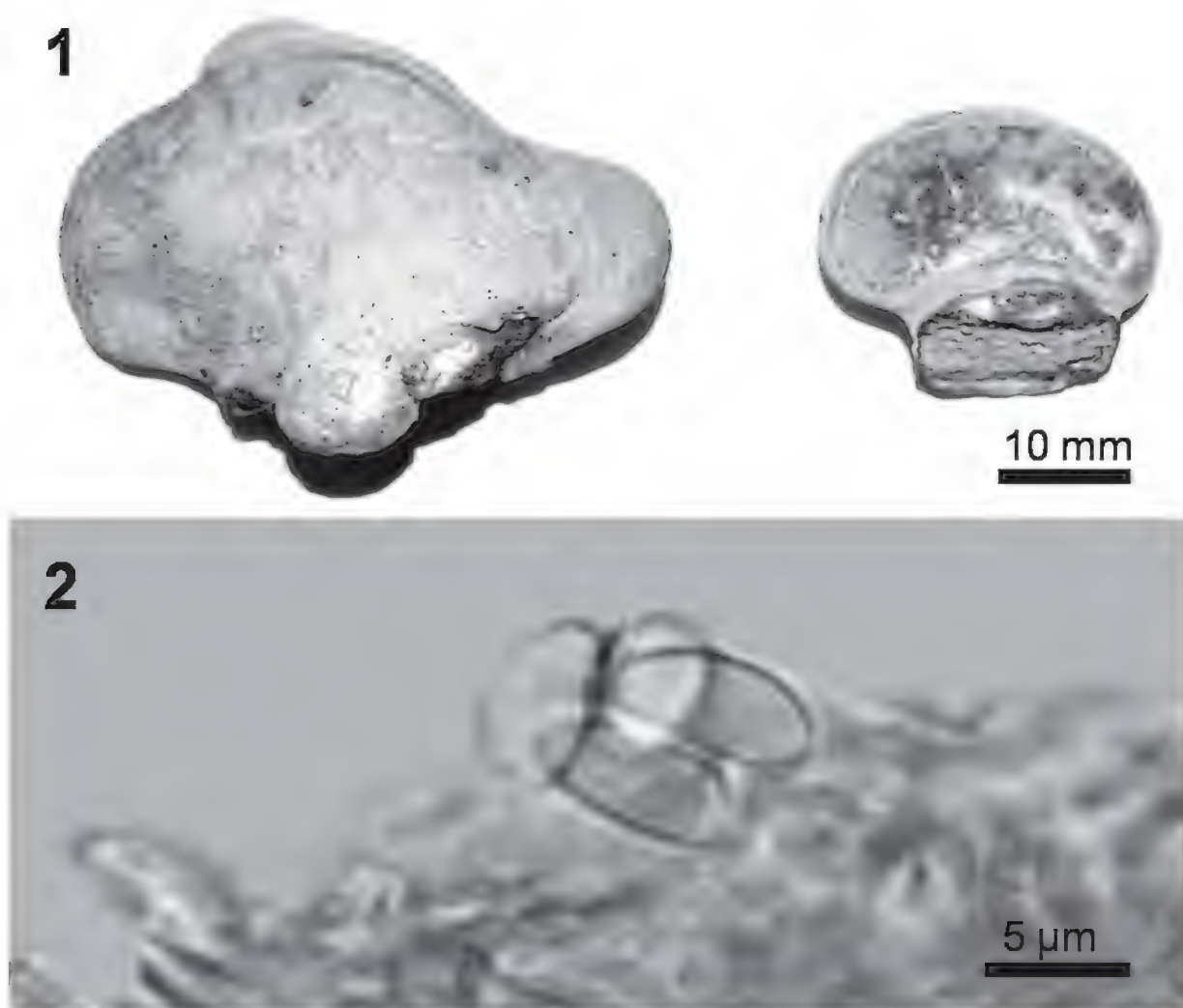
(INPA 82954, as *Polyporus modestus*); 1.VIII.1978, R. B. Singer & I. de J. Araújo 11342 (INPA 82956, as *Polyporus modestus*); 27.VI.1983, M.A. de Jesus 132 (INPA 183814); 29.VII.1983, M.A. de Jesus 125 (INPA 183808); 22.V.1985, M.A. de Jesus 735-746 (INPA 185965, INPA 185976); 6.X.1985, K.F. Rodrigues et al. 801 (INPA 137087); 22.VI.1989, R.E. Hanada 1006 (INPA 186282); 17.VII.1990, M.A. de Jesus 1448 (INPA 192694); 14.XII.1990, M.A. de Jesus 1433 (INPA 192686); 17.XII.1990, M.A. de Jesus 1455 (INPA 192700); 9.I.1992, R.E. Hanada 1522 (INPA 192731); 15.IX.1992, M.A. de Jesus 1533 (INPA 192739); 9.II.1993, M.A. de Jesus 1542 (INPA 192744); Manicoré, 14.IV.1985, K.F. Rodrigues et al. 126 (INPA 128926, as *Polystictus* sp.); Novo Aripuanã, 23.IV.1985, K.F. Rodrigues et al. 323 (INPA 128987); Presidente Figueiredo, 25.VI.1984, M.A. de Jesus 443 (INPA 185381); loc. n. det., 6.X.1966, G.T. Prance et al. 2602 (INPA 18770, as *Polyporus modestus*); 1.XI.1977, E. M. de L. Freire 145 (INPA 70048, as *Coriolopsis* sp.); 14.I.1978, I. de J. Araújo et al. 887 (INPA 78643, as *Polyporus* sp.); 21.I.1978, M. L. Farr et al. 176 (INPA 164405); 22.I.1978, M. L. Farr et al. 222 (INPA 164432); Pará: Itaituba, 29.IX.1977, M.A. de Sousa & L. F. Coêlho 19 (INPA 74690, as *Fomitopsis* sp.); 1.X.1977, M.A. de Sousa & L. F. Coêlho 82 (INPA 74528, as *Coriolopsis* sp.); 2.X.1977, M.A. de Sousa & L. F. Coêlho 105 (INPA 74534, as *Coriolopsis* sp.); 4.X.1977, M.A. de Sousa & L. F. Coêlho 19 (INPA 74627, as *Fomitopsis* sp.); Oriximinã, 17.VI.1980, V.L.R. Bononi 347 (INPA 103419, as *Polyporus* sp.); 19.VI.1980, V.L.R. Bononi 439 (INPA 103483); 29.VI.1980, V.L.R. Bononi 788 (INPA 103706); 1.VII.1980, V.L.R. Bononi 889 (INPA 103766, as *Coriolus* sp.); 2.VII.1980, V.L.R. Bononi 970 (INPA 103829, as *Coriolus* sp.); Melgaço, VII.2006, T. B. Gibertoni (URM 79929, URM 79928, URM 79931, URM 79934, URM 79927, URM 79930, URM 79932, URM 79933, URM 79935); VII.2007, T. B. Gibertoni (URM 79937, URM 79944, URM 79941, URM 79943, URM 79940, URM 79942, URM 79938, URM 79945, URM 79948, URM 79939, URM 79936); II.2008, T. B. Gibertoni (URM 79947, URM 79946); Rondônia: loc. n. det., 3.VII.1968, K.P. Dumont et al. 56-61 (INPA 65097, INPA 65101); Porto Velho, Parque Natural Municipal de Porto Velho, II.2007, A.C. Gomes-Silva 172-241 (URM 79217, URM 79572); VII.2007, A.C. Gomes-Silva 06-52-53-173-191-233-237 (URM 79570, URM 79571, URM 79216, URM 79218, URM 79221, URM 79219, URM 79220); II.2008, A.C. Gomes-Silva 285-318 (URM 79024, URM 79025); VII.2008, A.C. Gomes-Silva 613-619 (URM 79576, URM 79577); Estação Ecológica de Cuniã, VII.2007, A.C. Gomes-Silva 233-237-242 (URM 79219, URM 79220, URM 79222); VII.2008, A.C. Gomes-Silva 566-567-578 (URM 79573, URM 79574, URM 79575); Roraima: Alto Alegre, 10.VI.1986, E.S.S. da Silva et al. 465-412 (INPA 154936, INPA 154908); 12.VI.1986, K.F. Rodrigues et al. 948 (INPA 143328); 19.VI.1986, K.F. Rodrigues et al. 1052 (INPA 143400); Boa Vista, 21.XI.1977, L. de L. J. Aguiar et al. 701 (INPA 78420); 19.VII.1989, M.A. de Jesus 886 (INPA 186191); Caracaraí, 16.XI.1977, I. de J. Araújo et al. 461 (INPA 76988); loc. n. det., 13.I.1969, G.T. Prance et al. 9275 (INPA 26410); 17.I.1969, G.T. Prance et al. 9320 (INPA 26456); 6.II.1969, G.T. Prance et al. 9643 (INPA 26779); 24.III.1971, G.T. Prance et al. 11197 (INPA 29598, as *Coriolopsis* sp.).

DESCRIPTION — Gilbertson & Ryvarden (1987).

DISTRIBUTION — Pantropical (Núñez & Ryvarden 2001). In Brazil, it was recorded in the states of Bahia, Pernambuco, São Paulo (Baltazar & Gibertoni 2009), Acre, Amazonas, Pará, Rondônia, Roraima (Gomes-Silva & Gibertoni 2009), and Mato Grosso (Gibertoni & Drechsler-Santos 2010).

NOTES — The species may be confused with *T. supermodesta*, but is distinguished by the smaller pores (6–10 per mm) and basidiospores ( $4-6 \times 1.5-2 \mu\text{m}$ ).





FIGURES 1–2. *Trametes ochroflava*. 1. Basidiomata. 2. Basidiospores.

*Trametes ochroflava* Cooke, Grevillea 9(no. 49): 12 (1880).

FIGURES 1–2

MATERIAL EXAMINED: BRAZIL. Acre: loc. n. det., 17.X.1980, B. Lowy et al. 750 (INPA 100602, as *Polyporus* sp.); 22.X.1980, B. Lowy et al. 850 (INPA 100679, as *Polyporus* sp.); Amazonas: Humaitá, 25.XI.1966, G.T. Prance & J.F. Ramos 3316 (INPA 18891); loc. n. det., 20.IX.1977, M.A. de Sousa 330 (INPA 74749); Rondônia: loc. n. det., 2.VI.1984, R.D. Goos et al. 1760 (INPA 125259, as *Polyporus* sp.); Roraima: loc. n. det., 17.XI.1977, I. de J. Araújo et al. 570 (INPA 77559, as *Daedalea* sp.); 18.VII.1986, B. Lowy et al. 2208 (INPA 145485, as *Ganoderma* sp.); Pará: Oriximinã, 28.VI.1980, V.L.R. Bononi 679 (INPA 103633).

DESCRIPTION — Ryvarden (1988).

DISTRIBUTION — Known from Brazil (Ryvarden 1988). In Brazil, reported from the states of Bahia, Rio de Janeiro and Rio Grande do Sul (Baltazar & Gibertoni 2009). It is a new record for the Brazilian Amazonia.

NOTES — This species (FIG 1) resembles poroid specimens of *Lenzites elegans*, which are whitish and thinner. The basidiospores were not found in the type and not previously known (Ryvarden 1988), but a few were seen in INPA 18891 (FIG 2) and are cylindrical, hyaline, thin-walled,  $8\text{--}10 \times 4 \mu\text{m}$ .

***Trametes pavonia*** (Hook.) Ryvarden, Norw. Jl Bot. 19: 236 (1972), nom. illegit., non (Berk.) Fr. 1851.

≡ *Boletus pavonius* Hook., Syn. Pl. 1: 10 (1822).

MATERIAL EXAMINED: BRAZIL. Amazonas: Barcelos, 11.VII.1985, E.S.S. da Silva et al. 283 (INPA 153723); Manaus, 13.V.1977, M.A. de Sousa & I. de J. Araújo 88-325 (INPA 74662, INPA 74718); 28.I.1997, M.A. de Jesus 1912 (INPA 216449); 2.V.1997, A. Luis 2270 (INPA 192823); loc. n. det., 26.VI.1971, G.T. Prance et al. 13735 (INPA 31951, as *Coriolopsis* sp.); Rondônia: loc. n. det., 8.XI.1979, R. H. Petersen 445 (INPA 110933, as *Polyporus* sp.); Roraima: Alto Alegre, 21.VI.1986, K.F. Rodrigues et al. 1090 (INPA 143431); loc. n. det., 18.VI.1986, B. Lowy et al. 2169 (INPA 145449, as *Polyporus* sp.).

DESCRIPTION — Gilbertson & Ryvarden (1987).

DISTRIBUTION — Tropical America to northern Argentina (Gilbertson & Ryvarden 1987). In Brazil, reported from the states of Alagoas, Pernambuco and Santa Catarina (Baltazar & Gibertoni 2009). It is a new record for the Brazilian Amazonia.

NOTES — This species is similar to *T. membranacea*, but the flexible, concentrically zonate basidiomata distinguish *T. pavonia*.

***Trametes versicolor*** (L.) Lloyd, Mycol. Writ. 6: 1045 (1921).

≡ *Boletus versicolor* L., Sp. pl. 2: 1176 (1753).

MATERIAL EXAMINED: BRAZIL. Amazonas: Novo Aripuanã, 27.IV.1985, K.F. Rodrigues 388 (INPA 129004, as *Daedalea* sp.); loc. n. det., 12.VIII.1977, M.A. de Sousa 244 (INPA 74642, as *Coriolus* sp.).

DESCRIPTION — Núñez & Ryvarden (2001).

DISTRIBUTION — Cosmopolitan (Núñez & Ryvarden 2001). In Brazil, in Pará (Gomes-Silva & Gibertoni 2009), Bahia, Paraná, Rio Grande do Sul, São Paulo, Santa Catarina (Baltazar & Gibertoni 2009) and now in Amazonas.

NOTES — This species is characterized by the thin, tomentose, zonate basidiomata, also extremely variable in color.

***Trametes villosa*** (Sw.) Kreisel, Monografias, Ciências, Univ. Habana, Ser. 4, 16: 83 (1971).

≡ *Boletus villosus* Sw., Fl. Ind. Occid. 3: 1923 (1806).

MATERIAL EXAMINED: BRAZIL. Amazonas: Manaus, 6.III.1997, M.A. de Jesus 2060 (INPA 192826, as *T. menziesii*); 2.V.1997, M.A. de Jesus 2269 (INPA 192827, as *T. menziesii*); 1.VII.1997, M.A. de Jesus 2346 (INPA 192818); 21.X.1997, M.A. de Jesus 2501 (INPA 192820); Roraima: Alto Alegre, 12.VI.1986, K.F. Rodrigues et. al. 938 (INPA 143318); 16.VI.1986, K.F. Rodrigues et. al. 996 (INPA 143363); 17.VI.1986, E.S.S. da Silva et al. 483 (INPA 154950); 18.VI.1986, K.F. Rodrigues et. al. 1037 (INPA 143391); 21.VI.1986, K.F. Rodrigues et. al. 1071 (INPA 143415); Boa Vista, 20.VII.1989, M.A. de Jesus 920 (INPA 186221); loc. n. det., 24.VII.1974, G.T. Prance et al. 21368 (INPA 45343, as *Coriolus pinsitus*); 16.VI.1986, B. Lowy et al. 1227 (INPA 144553, as *Coriolus* sp.).

DESCRIPTION — Gilbertson & Ryvarden (1987).

DISTRIBUTION — Neotropical, also known from subtropical areas in the USA and Argentina (Gilbertson & Ryvarden 1987). In Brazil, recorded in the states of Amapá,

Pará, Roraima (Gomes-Silva & Gibertoni 2009), Bahia, Paraná, Rio de Janeiro, Rio Grande do Sul, São Paulo, Santa Catarina (Baltazar & Gibertoni 2009, Gibertoni & Drechsler-Santos 2010), and now in Amazonas.

NOTES— The thin basidiomata with large pores (2–3/mm) characterizes this species.

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We would like to thank Dr. Annarosa Bernicchia and Dr. Erast Parmasto for critically reviewing the manuscript; Ana Cristina R. Souza, curator of the HFSL, for support during the field trips of ACGS; Carlos Franciscan, curator of INPA, for the loan of exsiccates; and the staff of the ECFPn and of the MPEG for support during the field trips of TBG. Further, we acknowledge the Conselho Nacional de Desenvolvimento Científico (CNPq) for the master scholarship and Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (Capes) for the doctorate scholarship of ACGS; the Instituto Internacional de Educação do Brasil (IEB) and the Gordon and Betty Moore Foundation for the Scholarship of Studies on Amazonia Conservation (BECA) to ACGS and TBG; the Dottorato di Ricerca in Ecologia Sperimentale e Geobotanica (Università degli Studi di Pavia, Italy), the Pós-Graduação em Biologia de Fungos (UFPE, Brazil) and the Instituto Nacional de Ciência e Tecnologia - Herbário Virtual de Plantas e Fungos (CNPQ - 573883/2008-4) for partially financing this study.

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## MYCOTAXON

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***Leucoagaricus dacrytus***  
– a new species from New Jersey, U.S.A.

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**Abstract** — *Leucoagaricus dacrytus* (Agaricaceae) is described as new from an old-growth *Quercus rubra* forest in New Jersey, U.S.A. This is a relatively small, brown species exuding golden drops over its surface, with a pileus with cobwebby patches (a cutis-like pileus covering), narrowly clavate cheilocystidia, and oblong-amygdaloid spores. It is close to the European species *La. tener*, from which it differs in the slightly smaller spores and in nrITS sequences.

**Key words** — biodiversity, North America, taxonomy

### Introduction

Several unknown lepiotaceous fungi were discovered during long-term mycological research in the Oakmoss Mycological Preserve, a forest with old-growth *Quercus rubra* and many secondary trees (*Fagus*, *Betula*, *Cornus*, *Acer*, *Carya*, *Prunus*, and sassafras) in New Jersey; the specimens were sent to the first author for identification. One of them is a striking brown species with golden droplets on the surfaces of the basidiocarps. A literature search (starting with Lincoff 1991, Bessette et al. 1997, moving to Murrill 1914, Kauffman 1924, and Smith 1954, 1966) did not result in a fitting name. Here it is described as a new species, based on the morphology and the nrITS sequences, and it is compared with *Leucoagaricus tener* (P.D. Orton) Bon from Europe, and species with similar general morphology from other parts of North America and Europe.

### Material & methods

Macroscopic descriptions were based on the photos and notes provided by the second author. Standard methods for describing basidiocarps were applied, using the

terminology of Vellinga & Noordeloos (2001). Colour codes are according to the Online Auction Color Chart™, indicated by 'oac' before a number. Microscopical observations were made on dried material. The notation [115,6,5] indicates that measurements were made on 115 spores in six samples in five collections. At least 15 spores were measured per collection. The lamellar characters and spore shape and size were observed in Congo Red in 10% ammonia followed by ammonia only, and the pileus covering was observed in 10% ammonia. The following abbreviations are used: L for number of lamellae, l for number of lamellulae between two lamellae, avl for average length, avw for average width, Q for quotient of length and width, and avQ for average quotient. The abbreviation *L.* is used for *Lepiota*, *La.* for *Leucoagaricus* and *Lc.* for *Leucocoprinus*. All collections are in UC. Herbarium abbreviations are according to Holmgren & Holmgren (1998). The Latin description of the new species has been deposited in MycoBank.

DNA was extracted from dried material using a Qiagen DNeasy® Blood and Tissue kit (Qiagen, Valencia, CA, USA). The nrITS region was amplified with the ITS-1F/ITS-4 primer set with an MJ PTC-100™ thermocycler (Applied Biosystems, Foster City, CA, USA) under conditions previously described (Gardes & Bruns 1993). PCR products were cleaned using 0.5 µl of ExoSAP IT (USB Corp, Cleveland, OH, USA) per reaction and cycled at 37°C for 45 min, followed by 80°C for 15 min. Sequencing was performed using Big Dye chemistry and an ABI PRISM 3100 Genetic Analyzer (both from Applied Biosystems, Foster City, CA, USA). Sequences were edited and contigs assembled using Sequencher 4.2.2 (Gene Codes Corporation, Ann Arbor, MI, USA). Newly produced sequences were deposited in GenBank, and their accession numbers are listed with the collections and all accession numbers are given in FIG. 3. American *Leucoagaricus* species with brown to black cobwebby patches on the pileus surface were chosen for sequence comparisons, mainly from species in the *L. atrodisca* species complex in California, but also based on BLAST searches in GenBank (Altschul et al. 1990). The sequences were aligned with the program MAFFT version 6 (Katoh et al. 2002, Katoh & Toh 2008). For the phylogenetic analyses the Maximum Parsimony option in PAUP\* v4 (Swofford 2002) was used. *Chlorophyllum rachodes* (Vittad.) Vellinga and *Leucoagaricus americanus* (Peck) Vellinga were chosen as outgroup. The analyses were only performed to determine whether the sequences matched sequences of previously sequenced species and collections.

## Taxonomy

### *Leucoagaricus dacrytus* Vellinga, sp. nov.

FIGURES 1 & 2

MYCOBANK MB516742

*Leucagarico tenero similis, sporis nonnihil minoribus (i.e. 5.9–7.4 µm longis 2.9–4.1 µm latis, in medio 6.3–6.8 µm longis 3.5–3.9 µm latis) plus quam 50 basibus ceteris in ITS1 differt.*

HOLOTYPE: "U.S.A., New Jersey, Hunterdon Co., Lebanon, Oakmoss Mycological Preserve, R.B. Balsley, 11 Sept 2006 (UC)."

ETYMOLOGY—from the Greek δακρυτος, tearful, because of the presence of drops on the basidiocarps.

PILEUS 10–33 mm, when very young paraboloid to hemispherical with inflexed margin, expanding to plano-convex, and finally applanate with low and broad



FIG. 1. *Leucoagaricus dacrytus* — Habitus (from Balsley, 12 July 2006).

umbo, when young almost completely dull brown (oac702) except for a marginal, lighter zone, later only brown at umbo, and very light at margin, covered in small fibrillose-cobwebby patches to tufts, dense at centre, thinner at margin and there showing off-white background, on drying slightly sulcate for up to 5 mm at margin, covered with scattered drops, changing from golden yellow (oac856) to brown with age. LAMELLAE, L = 45–55, l = 1(–3), moderately crowded to crowded (2–3/1 mm, measured at pileus margin), free, relatively close to stipe, off-white to pale cream coloured, with concolourous not obviously cystidiose edge. STIPE 20–50 × 1.5–3.5 mm cylindrical and slightly wider, 3–5 mm, at utmost base, off-white above annulus, below annulus off-white changing to pale yellow with age, slightly darker when scratched, when fresh covered in pale yellow to yellow (oac856) drops, sometimes with basal white tomentum, hollow, with white mycelial cords at base. ANNULUS an ascending funnel with small flaring part, off white, with golden drops on underside. SMELL none.

BASIDIOSPORES [105,6,5] in side view 5.9–7.4 × 2.9–4.1  $\mu\text{m}$ ,  $\text{avl} \times \text{avw} = 6.3\text{--}6.8 \times 3.5\text{--}3.9 \mu\text{m}$ ,  $Q = 1.5\text{--}2.15$ ,  $\text{av}Q = 1.7\text{--}1.85$ , oblong-amygdaliform, with rounded apex, in frontal view oblong-ovate, with guttule, thick-walled some with a hint of an apical germ pore, young spores in particular congophilous, cyanophilous, dextrinoid, when young clearly metachromatic in Cresyl Blue, later less evidently so. BASIDIA 13.5–27 × 6.0–8.5  $\mu\text{m}$ , with 4 sterigmata, without clamp connection. LAMELLA EDGE sterile, with a band of cystidia. CHEILOCYSTIDIA 22–50 × 6–13  $\mu\text{m}$ , narrowly clavate to subutriform, some clavate, some slightly strangulated, subtly variable in shape, slightly thick-walled, and colourless. PLEUROCISTIDIA absent. PILEUS COVERING cutis-like, made up of strands of hyphae with up to 5 coloured elements in a row; terminal elements, slightly differentiated and wider than penultimate elements, 27–95



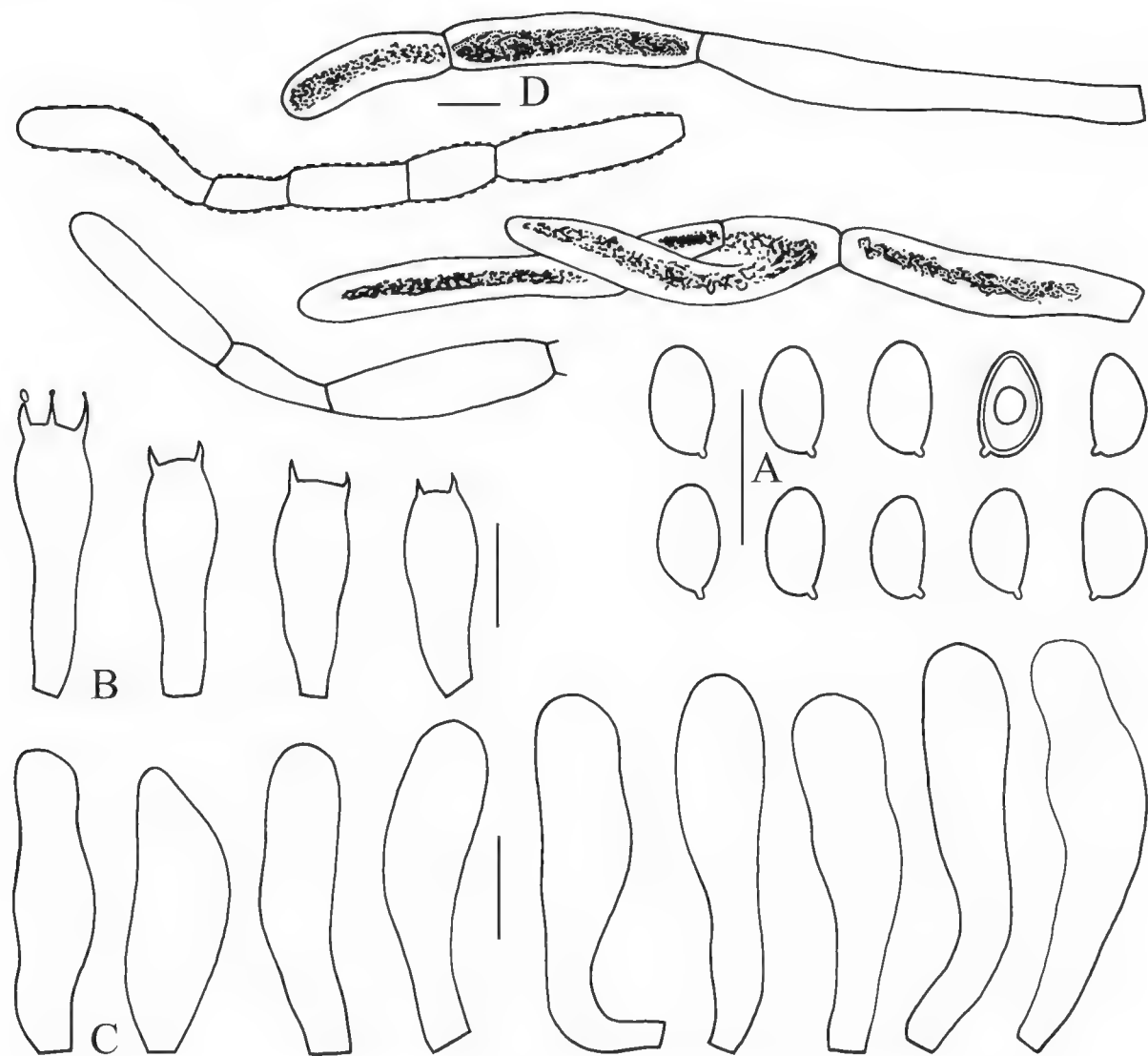


FIG. 2. *Leucoagaricus dacrytus* — A. spores, B. basidia, C. cheilocystidia, D. pileus covering elements (all from Balsley, 30 Aug. 2007). Scale bars are 10  $\mu$ m.

$\times$  5–12.5(-23)  $\mu$ m, with rounded apex; pigment brown, intracellular in big blob, or parietal and incrusting, in all elements, also the terminal ones. CLAMP CONNECTIONS absent.

**HABITAT & DISTRIBUTION** — In small groups, on decayed wood, most likely from *Quercus rubra*, in a deciduous forest (old growth *Quercus rubra* plus various other deciduous trees); so far only known from one spot at the type locality in New Jersey. July–Sept.

**COLLECTIONS EXAMINED**—U.S.A., New Jersey, Hunterdon Co., Lebanon, Oakmoss Mycological Preserve, 12 July 2006, R.B. Balsley (nrITS GU903308); ibidem, 8 Sept 2006, R.B. Balsley; ibidem, 11 Sept 2006, R.B. Balsley (nrITS GU903309)(Holotype, UC); ibidem, 26 Aug 2007, R.B. Balsley; ibidem, 30 Aug 2007, R.B. Balsley.

**Discussion**

*Leucoagaricus dacrytus* is characterized by brownish tinges in the pileus, the golden drops exuded on the basidiocarp surface, and microscopically by the

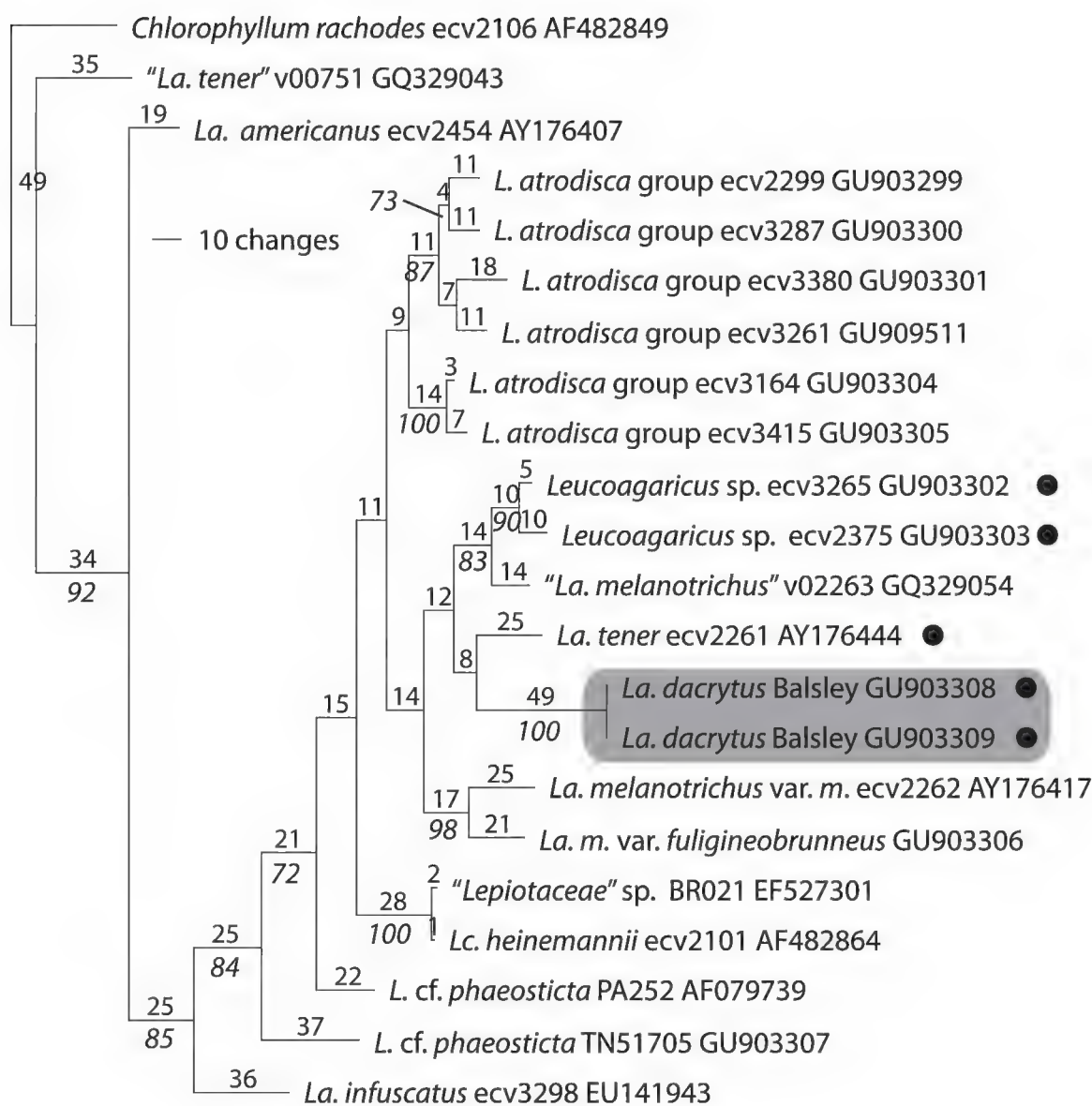


FIG. 3. Phylogram based on parsimony analyses of the nrITS region of a selection of *Leucoagaricus* and *Leucocoprinus* species with brown to black cobwebby scales made up of repent hyphae on the pileus surface. The one and only most parsimonious tree is presented, based on 211 informative characters. The numbers above branches refer to the number of changes, those below, in italics, to bootstrap values. The symbol ● indicates the presence of exudates. The newly described species *La. dacrytus* is highlighted. *Chlorophyllum rachodes* and *La. americanus* are outgroup taxa.

relatively small spores, the narrowly clavate cheilocystidia, and the cutis-like pileus covering with intracellular and incrusting pigments.

It is close, both morphologically and molecularly, to the European species *La. tener*, which differs in having smaller basidiocarps, slightly longer and wider spores, and more cylindrical cheilocystidia (Orton 1960, Uljé 1984, Vellinga 2001). These morphological differences are small and subtle. The nrITS sequences of both taxa are quite different (FIG. 3), with at least 50 different base pair positions in the ITS1 alone.

Drops on the basidiocarp surface are not restricted to these two species, but are found in many species in the *Leucoagaricus/Leucocoprinus* clade of the *Agaricaceae*, such as *Leucocoprinus cepistipes* (Sowerby) Pat. (sensu Lange 1935), and *Lc. lacrymans* T.K.A. Kumar & Manim. The former often has drops on its greyish pileus, stipe, and annulus, but the spores have a germ pore, and the cheilocystidia are big with an apical excrescence (e.g. Vellinga 2001). *Leucocoprinus lacrymans* from India also has spores with a germ pore, stains reddish when damaged, and has long, cylindrical cheilocystidia (Kumar & Manimohan 2004).

Neither the origin of the drops, nor the composition of them is known for this group of fungi. Many polypore species exude drops during the growing period, and it is known that *Pseudoinonotus dryadeus* (Pers.) T. Wagner & M. Fisch. exudates have a negative effect on the growth of gram positive bacteria (Blackwell & Adams 1985).

Other species that bear some resemblance to the presently described species are the following: *Leucoagaricus brunneocingulatus* (P.D. Orton) Bon, known from the United Kingdom (Orton 1960) and Italy (Migliozi & Perrone 1991), lacks drops, is red-brown on the pileus, and has a brown-rimmed annulus. The brown-scaled *La. brunneosquamulosus* P. Mohr & Dähncke, described from the Canary Islands, is clearly different because of the spores with a distinct germ pore and upright cylindrical to narrowly lageniform terminal elements in the pileus covering, and the narrow, cylindrical cheilocystidia (Mohr & Ludwig 2004). *Leucoagaricus infuscatus* Vellinga is another brown-squamulose species, with a cutis-like pileus covering; its brown pileus centre contrasts with the white background, and the absence of drops and the narrowly clavate to almost capitate cheilocystidia differentiate it from *La. dacrytus* (Vellinga 2007).

Sequences of species with comparable, but almost black pileus coverings, such as *L. atrodisca* Zeller, *La. melanotrichus* (Malençon & Bertault) Trimbach and *Lc. heinemannii* Migl., have been added to the group for comparison with *La. dacrytus* and *La. tener*, along with some unnamed species from California. Grey-brown species with drops on the basidiocarps are indicated in the resulting hypothetical phylogeny of FIG. 3. This group is species rich, with representatives all over the world, and desperately in need of morphological revision.

The species described and depicted by La Chiusa (1999) as *La. tener* is different from the original as described by Orton (1960); it lacks drops, the pileus covering is made up of velvety patches, not the cobwebby patches of *La. tener*, and its nrITS sequence (GenBank accession number GQ329043) differs significantly from the Dutch collection of *La. tener* (GenBank accession number AY176444; FIG. 3). Likewise, Migliozi & Coccia's interpretation of *La. tener* differs from Orton's concept (Migliozi & Coccia 1990, Vellinga 2001), though an nrITS sequence is not available for their specimens.

It is possible that *La. dacrytus* was described under a different name from North America, although the presence of droplets on the basidiocarps is not mentioned in any of the species descriptions, nor in the keys to the *Lepiota* species provided by Murrill (1914) and Kauffman (1924). Murrill included in his key all the species known at that time, and described by Peck, Morgan and others. The presence of droplets could have been noticed by the collector, but interpreted as unimportant or the result of external factors, such as rain. Reid (1995) did not report the presence of drops on *La. tener* basidiocarps, though Orton (1960) in the original description, did notice them. Recently, the presence of drops was omitted in the description of *L. furfuraceipes* Han C. Wang & Zhu L. Yang, a new species from Yunnan, China, and northern Thailand (Wang & Yang 2005). This species is common in northern Thailand, where the first author has repeatedly collected it; drops were always present on stipe and annulus, leaving dark spots on the annulus margin.

This paper hopefully will draw attention to the taxonomic significance of exudate drops on the basidiocarps of lepiotaceous fungi, and will also provide a stimulus to investigate this group of beautiful fungi in the eastern parts of North America.

### Acknowledgments

Jan Frits Veldkamp (Nationaal Herbarium, Leiden, the Netherlands) helped with the Latin diagnosis, and John Lennie edited the English. The article benefited from the reviews by Dr. Zai-Wei Ge and Dr. Nancy S. Weber. Funding by NSF grant DEB 0618293 for ECV is gratefully acknowledged.

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## MYCOTAXON

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Volume 113, pp. 81–85

July–September 2010

**A new pathogen of scale insects,  
*Aschersonia fusispora* sp. nov. (*Clavicipitaceae*)  
from Guangxi Province, China**

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**Abstract** — A new anamorphic species, *Aschersonia fusispora*, is described and illustrated based on collections from a natural forest in Guangxi Province of China. The species, which occurs on unidentified *Coccidae* larvae, is characterized with thinly pulvinate, slightly convex, pale orange stromata that are surrounded by a broad membranous hypothallus, wide ostiolar openings, and a 0.3–0.5 µm conidial width.

**Key words** — entomopathogenic fungus, taxonomy, new taxa

### Introduction

Species of the fungal genus *Aschersonia* Mont. (teleomorph *Hypocrella* Sacc.) parasitize scale insects (*Coccidae* and *Lecaniidae*, *Homoptera*) and whiteflies (*Aleyrodidae*, *Homoptera*) throughout tropical and (less often) subtropical regions, often resulting in epizootic events (Montagne 1848; Petch 1921; Mains 1959a,b; Chaverri et al. 2008; Mongkolsamrit et al. 2009; Qiu & Guan 2010). They are characterized by brightly colored pulvinate, subglobose or discoid stromata sometimes having a hypothallus, phialidic conidiogenous cells, the presence of pycnidial paraphyses, and unicellular, fusiform, and hyaline conidia which are produced in a mass of copious slime (Petch 1925; Mains 1959b; Chaverri et al. 2005, 2008).

During a survey on the biodiversity of insecticidal fungi in Guangxi province of China in 2008, two specimens of entomopathogenic *Aschersonia* were collected in evergreen broadleaved forests of the Huaping National Nature Reserve and the Maoershan National Nature Reserve. The general morphology

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of the specimens, such as flask-shaped pycnidia formed in stroma, slender branched conidiophores, fusoid conidia, and parasitism on homopteran insects, fit the generic concept of *Aschersonia*. The narrow and short-fusoid conidia, thinly pulvinate, slightly convex, pale orange stromata, and the presence of a broad membranous hypothallus differ from any described *Aschersonia* species.

### Materials and methods

Two collections from Guangxi province were studied. Conidiomata were carefully dissected with a razor blade and mounted in water or lactic acid mixed with cotton blue on a slide. The method of fungal measurements and microscopic features used in this study is the same as that described previously by Qiu et al. (2009). Colour names were described following Kornerup & Wanscher (1967). The voucher specimens studied were deposited in the Mycology Herbarium, Fujian Agricultural and Forestry University (MHFAFU).

### Taxonomy

*Aschersonia fusispora* Jun Z. Qiu, C.Y. Sun & Xiong Guan, sp. nov.      FIGS. 1A–F  
MYCOBANK MB 515186

*Stromata pulvinata, vel circularia, ex hyphis dense coactis composita, deorsum sparsa, hypothallum membranaceum ad 1.5 mm diam. 0.5 mm altum flavo-brunneum formantia, superficie aliquot orificiis ut punctis magnis visibilibus praedita. Pycnidialia, plerumque singula, in medio stromate immersa, 91–126 µm alta, 61–81 µm diam. Phialides cylindricae, ad 10 µm longae. Paraphyses pycnidiales praesentes, filiformes, flexuosae, ad 78 µm longae, 0.9 µm latae. Conidia fusioidea, utrinque rotundata 2.8–3.5 × 0.3–0.5 µm.*

TYPE — J.Z. Qiu, C.Y. Sun & X. Guan 367, MHFAFU 20837 (**holotype**) on *Coccidae*; Huaping National Nature Reserve, Guangxi Prov., Lingui County, Huaping, China, alt. 1600 m, 28.X.2008; J.Z. Qiu, C.Y. Sun & X. Guan 388, MHFAFU 20858 (**paratype**) on *Coccidae*; Maoershan National Nature Reserve, Guangxi Prov., Longsheng County, Maoershan, China, alt. 1200 m, 29.X.2008.

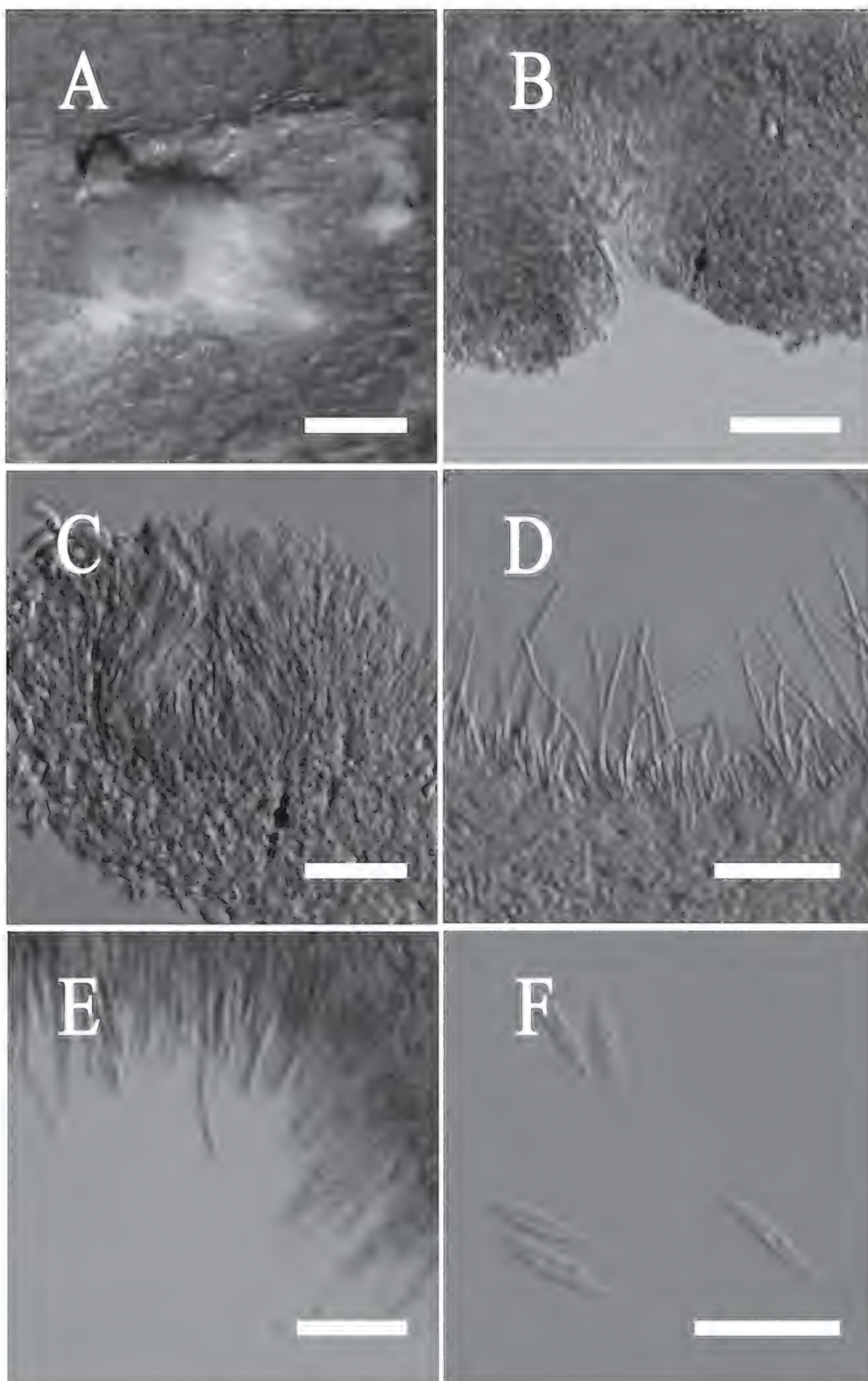
ETYMOLOGY — Refers to the fusiform conidia of this species.

TELEOMORPH: None known.

STROMATA thinly pulvinate, circular, slightly convex, consisting of dense hyphae, base spreading, forming a brownish-yellow membranous hypothallus up to 1.5 mm diam., 0.5 mm high, pale orange when fresh, several ostiolar openings as large dots visible on the surface. PYCNIDIA usually single, embedded in the centre of the stroma, 91–126 µm high, 61–81 µm diam. Conidiogenous cells phialidic, cylindrical, up to 10 µm long. PARAPHYSES present, linear, filiform,

FIG.1 *Aschersonia fusispora*. A: Stroma; B: Pycnidium; C: Longitudinal section of a flask-shaped pycnidium; D: Paraphyses; E: Conidiophores and conidiogenous cells; F: Conidia.

Scale bars: A = 1 mm; B,C,D = 50 µm; E = 20 µm; F = 5 µm.





flexuous, up to 78  $\mu\text{m}$  long, 0.9  $\mu\text{m}$  wide. CONIDIA fusoid, sometimes narrowly fusiform, with rounded ends,  $2.8\text{--}3.5 \times 0.3\text{--}0.5 \mu\text{m}$ .

COMMENTS—*Aschersonia fusispora* is characterized by the pale orange, thinly pulvinate, small stromata, the small conidia, the wide ostiolar openings, and the presence of paraphyses and hypothallus. Two previously described species of *Aschersonia*, *A. microspora* Sacc. and *A. minutispora* Hywel-Jones & Mongkolsamrit (Petch 1921, Mains 1959a,b, Hywel-Jones & Evans 1993, Chaverri et al. 2005, 2008, Mongkolsamrit et al. 2009) also have spores of the similar size. However, *A. microspora* differs in having pale brown stromata consisting of dense interwoven hyphae and globose or narrowly oval and wider conidia ( $2\text{--}4 \times 1.5 \mu\text{m}$ ), and lacking pycnidia. *A. minutispora* differs in possessing larger cream-brown stromata (2.5 mm in diam., 2 mm high), more voluminous pycnidia (350–400  $\mu\text{m}$  high, 300–350  $\mu\text{m}$  in diam.), bigger conidia ( $5\text{--}6 \times 1.2\text{--}1.5 \mu\text{m}$ ), and longer pycnidial paraphyses up to 150  $\mu\text{m}$  long and 1.5  $\mu\text{m}$  in width.

### Acknowledgements

The authors are grateful to Prof. Guo-Zhong Lv (Centre for Bioresources and Environment, Dalian Nationalities University) and Dr. Ryan Kepler (Department of Botany and Plant Pathology, Oregon State University) for serving as pre-submission reviewers. We also express our deep thanks to Prof. Jian-Yun Zhuang (Institute of Microbiology, Chinese Academy of Sciences) for assistance in correction of the Latin diagnoses, and to Dr. Shaun Pennycook for nomenclatural review. This project was financed by a general grant (30500005) from the National Natural Science Foundation of China, the Programs for Science and Technology (2007F5022) and the Educational Programs for Science and Technology Development (JA09085), operated by Fujian Provincial Department of Education, and the Key Project (2006S0002) from Fujian Provincial Department of Science and Technology.

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## MYCOTAXON

DOI: 10.5248/113.87

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**Two new species of *Septobasidium* (Septobasidiaceae)  
and *S. pallidum* new to China**CHUNXIA LU<sup>1,2</sup> & LIN GUO<sup>1\*</sup>*Ch.x.lu@hotmail.com & \*guol@im.ac.cn*<sup>1</sup>*Key Laboratory of Systematic Mycology and Lichenology  
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**Abstract** — Two new species, *Septobasidium meridionale* on *Litsea cubeba* associated with *Aulacaspis* sp. and *S. aulacaspidis* on an unidentified tree associated with *Aulacaspis* sp., are described. *Septobasidium pallidum* on *Zanthoxylum bungeanum*, *Z. simulans*, and *Pyrus phaeocarpa* is new to China.

**Key words** — *Pucciniomycetes*, *Septobasidiales*, taxonomy

Previously, a new species of *Septobasidium* was found in Hainan province (Lu & Guo 2009a). In December 2009, many specimens of *Septobasidium* were collected from the same area. Among them, an additional two new species are described as follows:

***Septobasidium meridionale* C.X. Lu & L. Guo, sp. nov.**

FIGS. 1–7

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*Basidiomata resupinata*, 4–8.5 cm longa, 1–6 cm lata, alba vel brunnea, margine determinata, superficie laevia vel volutina, in sectione 840–1000 µm crassa. Subiculum brunneum vel hyalinum, 20–50 µm crassum. Columnae hyalinae vel brunneolae, 40–130 µm altae, 40–340 µm latae, ex hyphis 3–4 µm latis compositae, superne ramosae tunc strato hypharum 580–780 µm alto formatae, interdum strata horizontalia formantes. Hymenium 40–90 µm crassum, hyalinum vel brunneum. Basidia cylindrica, recta vel curvata, 4-cellularia, 27–36 × 7–9.5 µm, hyalina vel brunneola. Sine probasidio. Basidiosporae non visae. Haustoria ex hyphis irregulariter spiralibus constantia.

TYPE: On *Litsea cubeba* Pers. (*Lauraceae*): China, Hainan, Bawangling, Nanchahe, alt. 600 m, 11.XII.2009, Y.F. Zhu & L. Guo 128, HMAS 240076 (**holotype**), associated with *Aulacaspis* sp. (*Diaspididae*).

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\*corresponding author



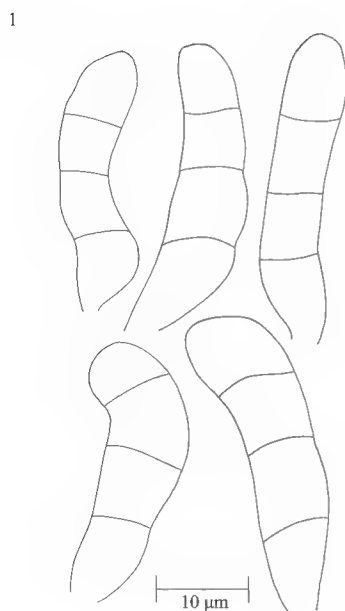


FIG. 1. Basidia of *Septobasidium meridionale* (HMAS 240076, holotype).

Basidiomata on trunks and branches, resupinate, perennial, 4–8.5 cm long, 1–6 cm wide, white or brown; margin determinate; surface smooth or velutinous. In section 840–1000  $\mu\text{m}$  thick. Subiculum 20–50  $\mu\text{m}$  thick, brown or hyaline. Pillars hyaline or brownish, 40–130  $\mu\text{m}$  high, 40–340  $\mu\text{m}$  wide, hyphae of pillars 3–4  $\mu\text{m}$  thick. Hyphal layer 580–780  $\mu\text{m}$  high, sometimes forming a horizontal layer. Hymenium 40–90  $\mu\text{m}$  thick, hyaline or brown. Basidia arising directly from the hyphae, cylindrical, straight or curved, 4-celled,  $27\text{--}36 \times 7\text{--}9.5$   $\mu\text{m}$ , hyaline or brownish, without a probasidial cell. Basidiospores not seen. Haustoria consisting of irregularly coiled hyphae.

REMARKS: Morphologically, *Septobasidium meridionale* is similar to *S. septobasidioides* (Henn.) Höhn. & Litsch., from which it differs in having short pillars (40–130  $\mu\text{m}$  vs 350–450  $\mu\text{m}$ ) and smaller basidia ( $27\text{--}36 \times 7\text{--}9.5$   $\mu\text{m}$  vs  $40\text{--}55 \times 8.4\text{--}10$   $\mu\text{m}$ ), and sometimes forming a horizontal layer.

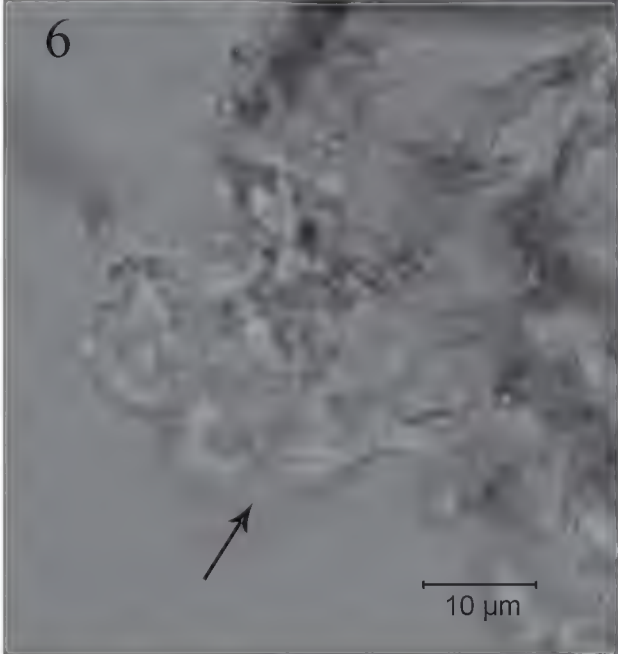
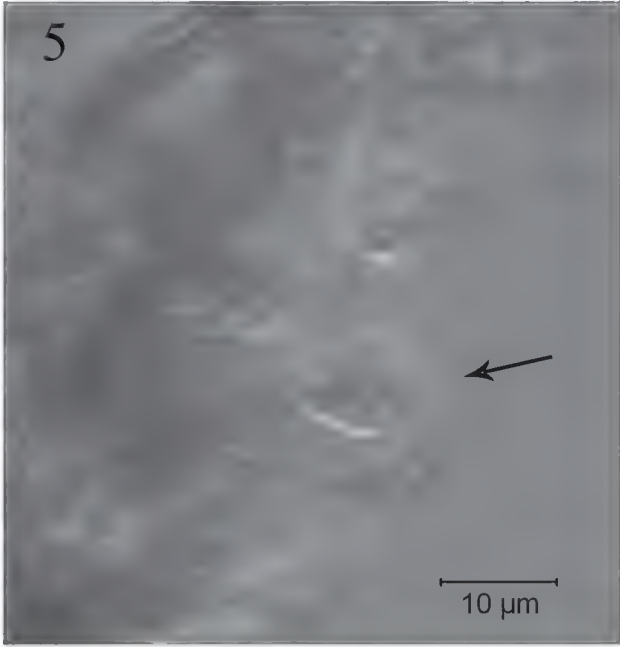
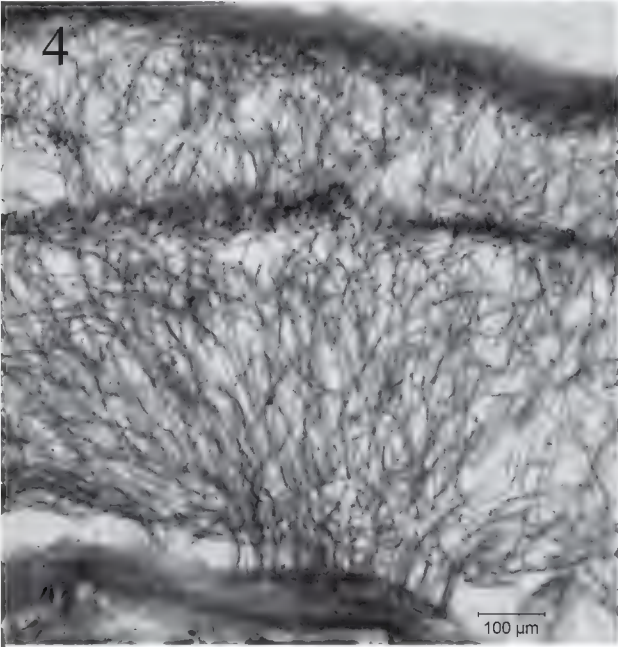
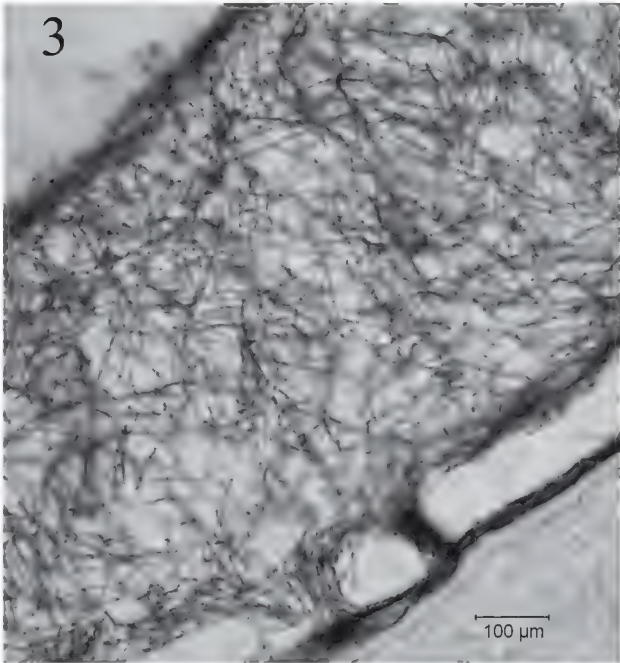
***Septobasidium aulacaspidis* C.X. Lu & L. Guo, sp. nov.**

FIGS. 8–12

MYCOBANK MB 518061

*Basidiomata resupinata, 0.2–7 cm longa, 0.1–5 cm lata, alba vel cinnamomeo-brunnea, margine indeterminata, superficie laevia vel velutina, in sectione 360–550  $\mu\text{m}$  crassa. Subiculum brunneum, 30–50  $\mu\text{m}$  crassum. Columnae hyalinae vel brunneolae, 200–260  $\mu\text{m}$  altae, 20–120  $\mu\text{m}$  latae. Hymenium 50–80  $\mu\text{m}$  crassum. Basidia cylindrica, recta vel curvata, 4-cellularia,  $28\text{--}50 \times 5\text{--}10$   $\mu\text{m}$ , hyalina vel brunnea. Basidiosporae ovoideae vel reniformes,  $10\text{--}16 \times 4\text{--}5.5$   $\mu\text{m}$ . Sine probasidio. Haustoria ex hyphis irregulariter spiralibus constantia.*

FIGS. 2–7. *Septobasidium meridionale* (HMAS 240076, holotype). 2. Basidiomata on trunk. 3–4. Sections of basidiomata. 5–6. Basidia (arrows). 7. Haustoria.



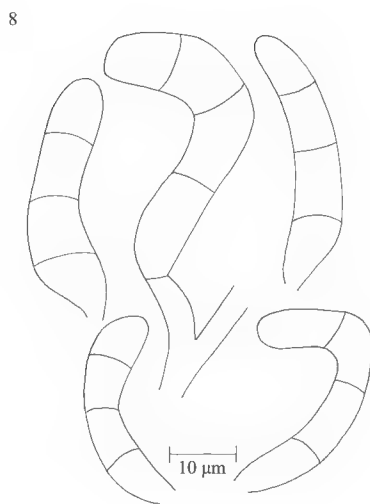


FIG. 8. Basidia of *Septobasidium aulacaspidis* (HMAS 240074, holotype).

TYPE: On unidentified tree [probably *Neolitsea* sp. (*Lauraceae*)]: China, Hainan, Jianfengling, alt. 900 m, 11.XII.2009, S.H. He 2803, HMAS 240074 (**holotype**), associated with *Aulacaspis* sp. (*Diaspididae*).

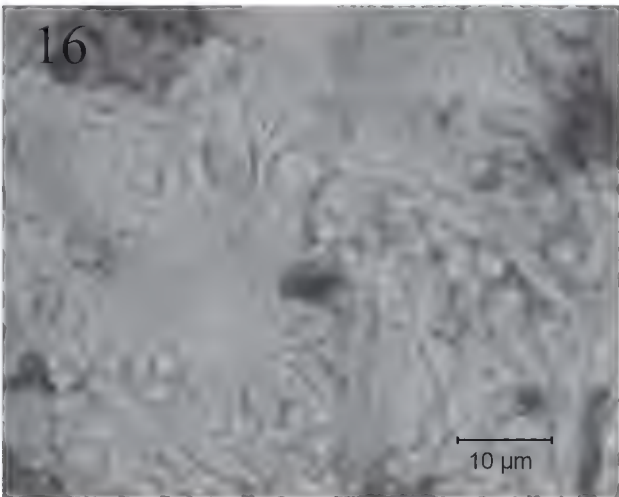
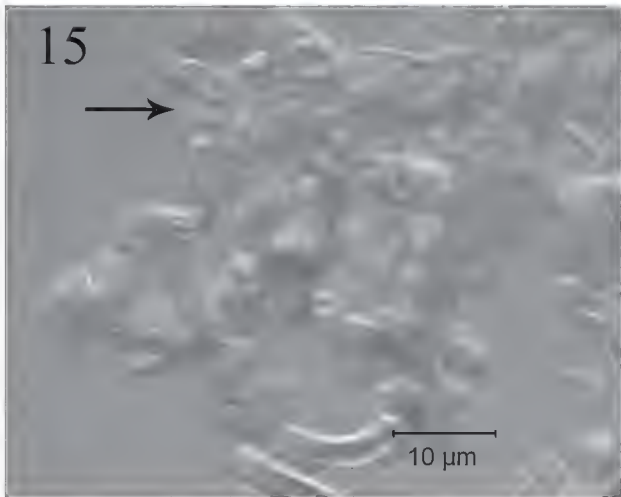
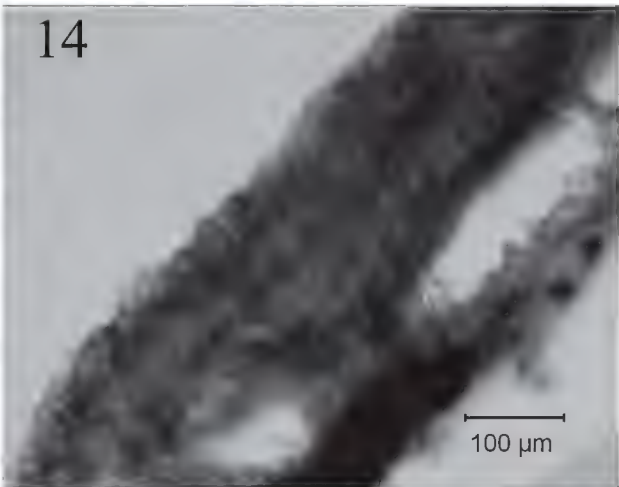
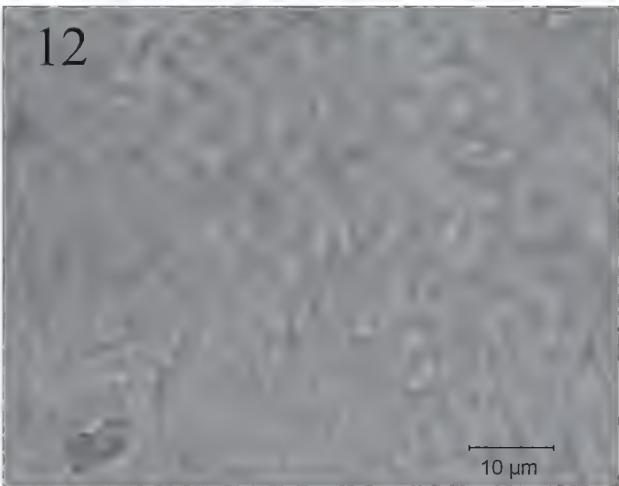
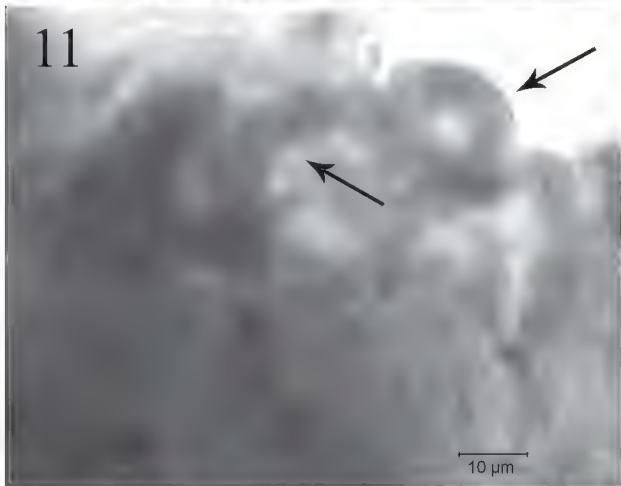
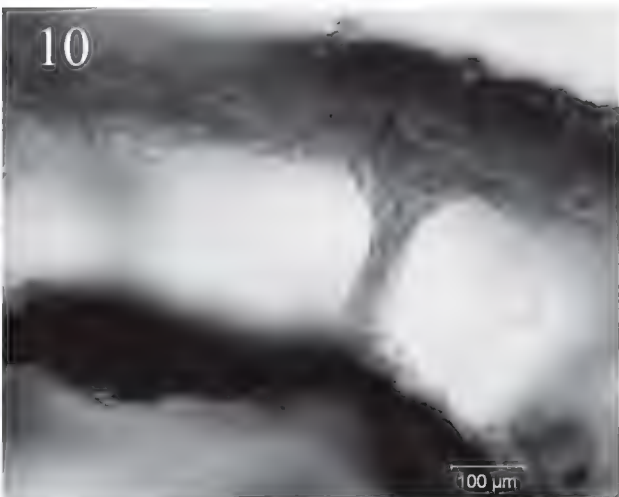
Basidiomata on trunks, resupinate, subcircular or irregular, often confluent 0.2–7 cm long, 0.1–5 cm wide, white or cinnamon-brown; margin indeterminate; surface smooth or velvety. In section 360–550  $\mu\text{m}$  thick. Subiculum 30–50  $\mu\text{m}$  thick, brown. Pillars 200–260  $\mu\text{m}$  high, 20–120  $\mu\text{m}$  wide, hyaline or brownish. Hymenium 50–80  $\mu\text{m}$  thick, with irregularly arranged upright hymenial hyphae. Basidia arising directly from the hyphae, cylindrical, straight or curved, 4-celled,  $28\text{--}50 \times 5\text{--}10 \mu\text{m}$ , hyaline or brown, without a probasidial cell. Sterigmata conical,  $5\text{--}13 \times 2 \mu\text{m}$ . Basidiospores ovoid or reniform,  $10\text{--}16 \times 4\text{--}5.5 \mu\text{m}$ , pale yellowish brown. Haustoria consisting of irregularly coiled hyphae.

REMARKS: *Septobasidium aulacaspidis* is similar to *S. pallidum*, but differs mainly in having indeterminate margin, smooth and velvety surfaces of basidiomata, and tall pillars (200–260  $\mu\text{m}$  vs 84  $\mu\text{m}$ ). *Septobasidium pallidum* has determinate margin, non-velvety surface of basidioma, and short pillars.

Recently, several specimens of a *Septobasidium* sp. on *Zanthoxylum bungeanum* and *Pyrus phaeocarpa* were collected in Sichuan province. They were identical to a specimen of *Septobasidium* sp. on *Zanthoxylum simulans* previously deposited in our herbarium. No basidia were found in the specimen. The fungus is identified as *S. pallidum*, a species unrecorded previously in China:

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FIGS. 9–12. *Septobasidium aulacaspidis* (HMAS 240074, holotype). 9. Basidiomata on trunk. 10. Section of basidioma. 11. Basidia (arrows). 12. Haustoria. FIGS. 13–16. *Septobasidium pallidum* (HMAS 199578). 13. Basidiomata on trunk. 14. Section of basidioma. 15. Basidium (arrow). 16. Haustoria.





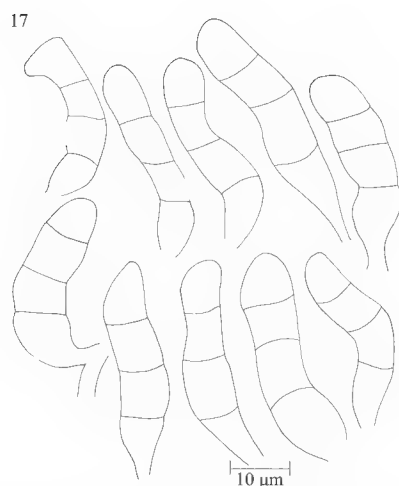


FIG. 17. Basidia of *Septobasidium pallidum* (HMAS 199578).

*Septobasidium pallidum* Couch ex L.D. Gómez & Henk, Lankesteriana 4(1): 88, 2004.

FIGS. 13–17

Basidiomata on trunks and branches, resupinate, subcircular, 0.2–7 cm long, 0.2–3 cm wide, patches frequently confluent, tilleul buff, yellowish-brown or brown; surface often smooth, with mounds and wrinkles, sometimes cracked; margin determinate, white. In section 220–510(–720) µm thick. Subiculum 20–60 µm thick. Pillars 40–90 µm high, 20–140 µm thick. Hyphal layer 100–400(–600) µm high. Hymenium 50–100 µm thick. Basidia arising directly from the hyphae without a probasidial cell, cylindrical, straight or slightly curved, 4-celled, 17–38(–42) × 6–12 µm, hyaline or brown. Sterigmata 12–27 µm long, 2–3 µm wide. Hymenial hyphae irregularly arranged. Basidiospores not seen. Haustoria consisting of irregularly coiled hyphae.

SPECIMENS EXAMINED: On *Zanthoxylum bungeanum* Maxim. (*Rutaceae*): China, Sichuan, Jinyang, alt. 1100 m, 15.VIII.2009, S.H. He, C.X. Lu, Y.F. Zhu & L. Guo 2680, HMAS 199578; Jinyang, alt. 600 m, 15.VIII.2009, S.H. He, C.X. Lu, Y.F. Zhu & L. Guo 2686, HMAS 196491; Jinyang, Wuke, alt. 1100 m, 15.VIII.2009, S.H. He, C.X. Lu, Y.F. Zhu & L. Guo 2681, HMAS 199582; Jinyang, Mufu, alt. 1600 m, 15.VIII.2009, S.H. He, C.X. Lu, Y.F. Zhu & L. Guo 2685, HMAS 196487; Mianning, Manshuiwan, Ganghe, alt. 1740 m, 24.VIII.2009, S.H. He, C.X. Lu, Y.F. Zhu & L. Guo 2789, HMAS 196493; Xide, Mianshan, Dengxiangying, alt. 2300 m, 23.VIII.2009, S.H. He, C.X. Lu, Y.F. Zhu & L. Guo 2781, HMAS 196492.

On *Zanthoxylum simulans* Hance (*Rutaceae*): China, Sichuan, Hanyuan, 14.XII.1937, Y. Hu, HMAS 10165.

On *Pyrus phaeocarpa* Rehder (*Rosaceae*): China, Sichuan, Xide, Tanshan, alt. 1860 m, 23.VIII.2009, S.H. He, C.X. Lu, Y.F. Zhu & L. Guo 2784, HMAS 199628.

To date, 26 species of *Septobasidium* have been reported in China (Sawada 1933, Couch 1938, Teng 1963, Tai 1979, Kirschner & Chen 2007, Lu & Guo 2009a,b,c, 2010, Lu et al. 2010) including the three species reported in this paper.

## Acknowledgements

The authors would like to express their deep thanks to Dr Eric H.C. McKenzie (Auckland, New Zealand) for serving as pre-submission reviewer, to Dr Shuanghui He (Beijing Forestry University) for serving as pre-submission reviewer and sending a specimen, to Prof. Jianyun Zhuang (Institute of Microbiology, Chinese Academy of Sciences) for Latin corrections, to Mr Ziyu Cao (Institute of Botany, Chinese Academy of Sciences) for identifying the host plants, to Prof. Sanan Wu (Beijing Forestry University) for identifying the scale insects, and to Mrs Xiangfei Zhu for inking in line drawings. This study was supported by the National Natural Science Foundation of China (No. 30499340 and No. 30870016) and the Ministry of Science and Technology of the People's Republic of China (No. 2006FY110500–5).

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## MYCOTAXON

DOI: 10.5248/113.95

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July–September 2010

**Taxonomic studies of *Helminthosporium* from China 5.  
Two new species from Hunan and Sichuan Province\***MENG ZHANG<sup>1</sup>, HAI-YAN WU<sup>2</sup> & ZHEN-YUE WANG<sup>1</sup>

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**Abstract** — Two new species of the genus *Helminthosporium* are reported from China: *H. bambusicola* and *H. hunanense*. Type specimens are deposited in the Herbarium of Henan Agricultural University: Fungi (HHAUF).

**Key words** — systematics, hyphomycetes, saprobes

In the course of a survey of *Helminthosporium* species in China, our previous research revealed eleven new species and five new records from China (Zhang et al. 2003, 2004, 2007, 2009). In this paper we describe two new species of this genus. Specimens studied are deposited in the Herbarium of Henan Agricultural University: Fungi (HHAUF).

***Helminthosporium bambusicola* Meng Zhang, H.Y. Wu & Zhen Y. Wang, sp. nov.**

MYCOBANK MB 516715

FIG. 1

*In substrato naturali coloniae effusae, atrae, pilosae. Mycelium plerumque immersum. Stromata partim superficialia et, partim immersa, atrobrunnea, pseudoparenchymatica, 10 µm alta, 25 µm lata. Conidiophora singularia spice lateribusque hypharum vel fasciculata ex stromata quoque, oriunda, simplicia, cylindrica, recta vel flexuosa, septata, levia, brunnea, interdum apicem versus pallidiora, 55–247 µm longa, 4–6 µm crassa, poris conidiiferis ad apicem et infra 1–2 septa supera praedita. Conidia obclavata, recta vel flexuosa, levia, pallide brunnea, 5–8-distoseptata, 36–66 µm longa, 6–11 µm crassa, apicem versus ad 2.0–4.5 µm gradatim attenuata.*

HOLOTYPE: ON DEAD *BAMBUSA* SP. CULM, Sichuan, China, 16. VIII 2008, coll. Z.Y. Wang, HHAUF<sub>08</sub> 0266.

ETYMOLOGY: Named for the substrate.

\* Supported by The National Natural Science Foundation of China (30870018 and 30970016)



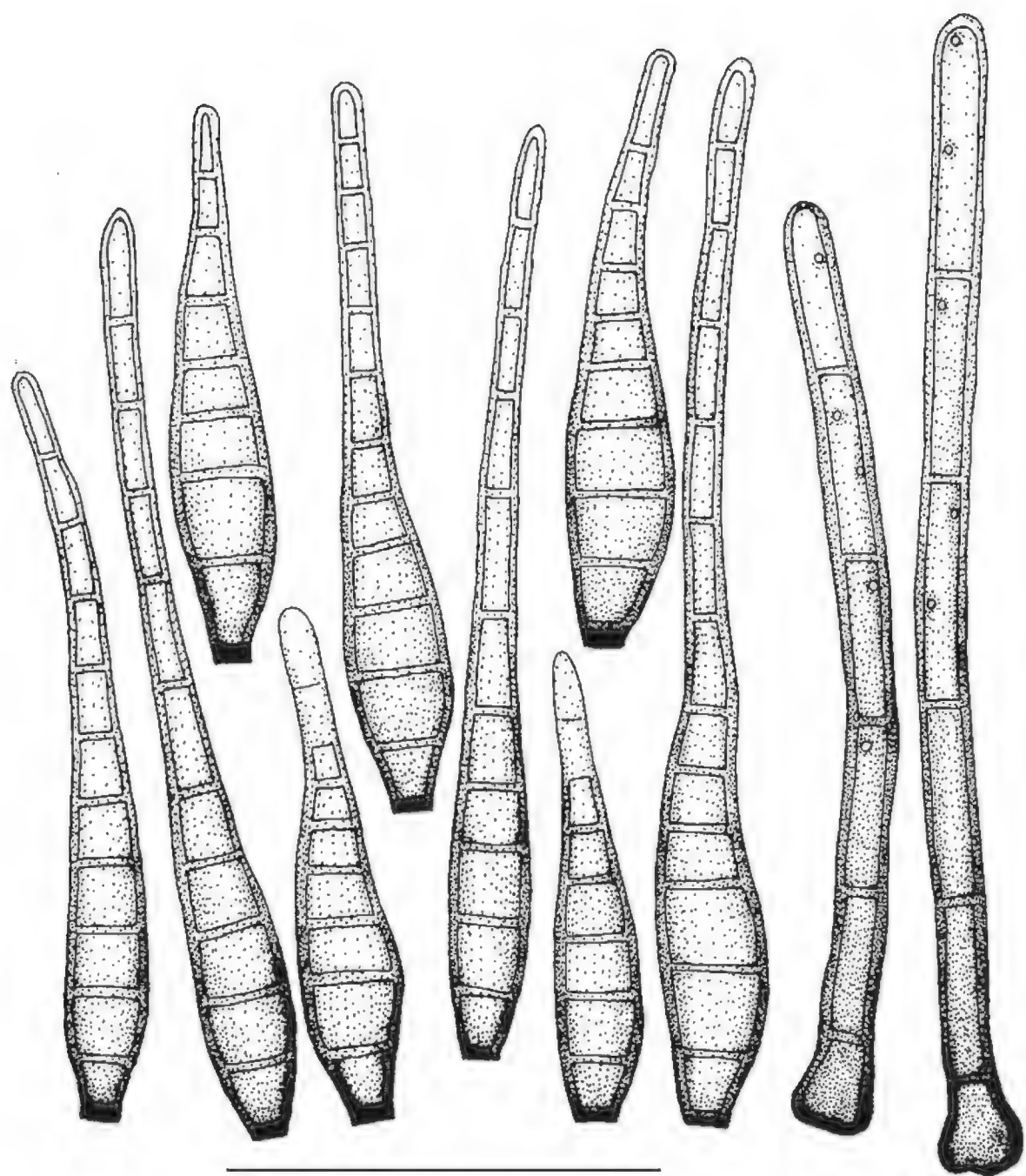


FIG. 1 *Helminthosporium bambusicola* (ex holotype, bar = 50  $\mu$ m)  
Conidia and conidiophores on natural substratum

Colony effused, black, hairy in the substrata. Mycelium mostly immersed in the substrata. Stromata partly superficial, partly immersed in the substrata, dark brown, pseudoparenchymatous, up to 10  $\mu$ m tall, 25  $\mu$ m wide. Conidiophores arising in fascicles from the upper cells of the stromata or solitary from the swelled cell of the mycelium, simple, cylindrical, straight or flexuous, thick-walled, smooth, brown, paler towards the apex, 55–247  $\mu$ m long, 4–6  $\mu$ m wide, with well-defined small pores (conidiogenous loci) at the apex and laterally just beneath the upper 1–2 septa. Conidia straight or slightly flexuous, obclavate, thin-walled 1–1.5  $\mu$ m thick, smooth, pale brown, paler towards the apex, 5–8–

distoseptate, 36–66  $\mu\text{m}$  long, 6–11  $\mu\text{m}$  wide, narrowing towards the apex to 2–4.5  $\mu\text{m}$  wide, scar not distinct at the base.

COMMENTS: Cooke (1892) published *Helminthosporium bambusae* on *Bambusa spinosa* Roxb. ex Buch.-Ham., a species distinguished by fewer distosepta (3–5) and slightly larger (60–70  $\mu\text{m}$  long, 12  $\mu\text{m}$  wide) conidia with slightly thinner ( $\leq 1.5$   $\mu\text{m}$ ) walls. Although the two species inhabit identical substrates, we feel that the morphological differences, while minor, support naming a new species. The new taxon also resembles *H. solani* Durieu & Mont. (Ellis 1961) in its conidial shape and size. However, *H. solani* is a pathogen on solanaceous hosts, has larger conidiophores (120–600  $\mu\text{m}$  long, 9–15  $\mu\text{m}$  wide at the base, 6–9  $\mu\text{m}$  wide at the apex), and slightly thicker ( $\geq 2$   $\mu\text{m}$ ) conidial walls.

***Helminthosporium hunanense*** Meng Zhang, H.Y. Wu & Zhen Y. Wang, sp. nov.

MYCOBANK MB 516716

FIG. 2

*In substrato naturali coloniae effusae, atrae, pilosae. Mycelium plerumque immersum. Stromata nulla. Conidiophora singularia vel 2–3 fasciculata ex spice lateribusque hypharum oriunda, recta vel flexuosa, cylindrica, septata, levia, brunnea vel atrobrunnea, interdum apicem versus pallidiora, 70–226  $\mu\text{m}$  longa, 5–7  $\mu\text{m}$  crassa, poris conidiiferis ad apicem et infra 1–3 septa supera praedita. Conidia obclavata, recta vel curvata, levia, moderate brunnea, apicem versus pallidiora, 4–12-distoseptata, 56–127  $\mu\text{m}$  longa, 10–14  $\mu\text{m}$  crassa, apicem versus ad 2–4  $\mu\text{m}$  gradatim attenuata, basi cicatrice majuscula fusca praedita.*

HOLOTYPE: On dead branches of an unidentified tree, Zhangjiajie, Hunan, China, 2 IX 2009, coll. M. Zhang, HHAUF<sub>09</sub> 0451.

ETYMOLOGY: Named for the collection locality (province).

Colony effused, black, hairy in the substrata. Mycelium mostly immersed in the substratum. Stromata absent. Conidiophores arising solitary or in fascicles from the cells of the mycelium, simple, straight or flexuous, septa at 17–31  $\mu\text{m}$  intervals, thick-walled, cylindrical, smooth, brown, 70–226  $\mu\text{m}$  long, 5–7  $\mu\text{m}$  wide just above the base cell which 8.5–14  $\mu\text{m}$  wide, with well-defined small pores at the apex and laterally beneath the upper 1–3 septa. Conidia obclavate, straight or curved, thin-walled, 1–1.5  $\mu\text{m}$  thick, smooth, middle brown, paler towards the apex, 4–12-distoseptate, 56–127  $\mu\text{m}$  long, 10–14  $\mu\text{m}$  wide in the widest part, narrowing towards the apex to 2–4  $\mu\text{m}$  wide, with a blackish-brown scar at the base, 1.5  $\mu\text{m}$  thick.

COMMENTS: The new species is most closely related to *Helminthosporium dalbergiae* M.B. Ellis in conidial morphology (shape and size). *H. dalbergiae* differs from this fungus by its much larger (300–550  $\mu\text{m}$  long, 10–12  $\mu\text{m}$  wide) conidiophores that arise from stromata and thicker ( $\geq 2$   $\mu\text{m}$ ) conidial walls.

The thinner conidial wall may be helpful in distinguishing both *H. hunanense* and *H. bambusicola* from other *Helminthosporium* species.

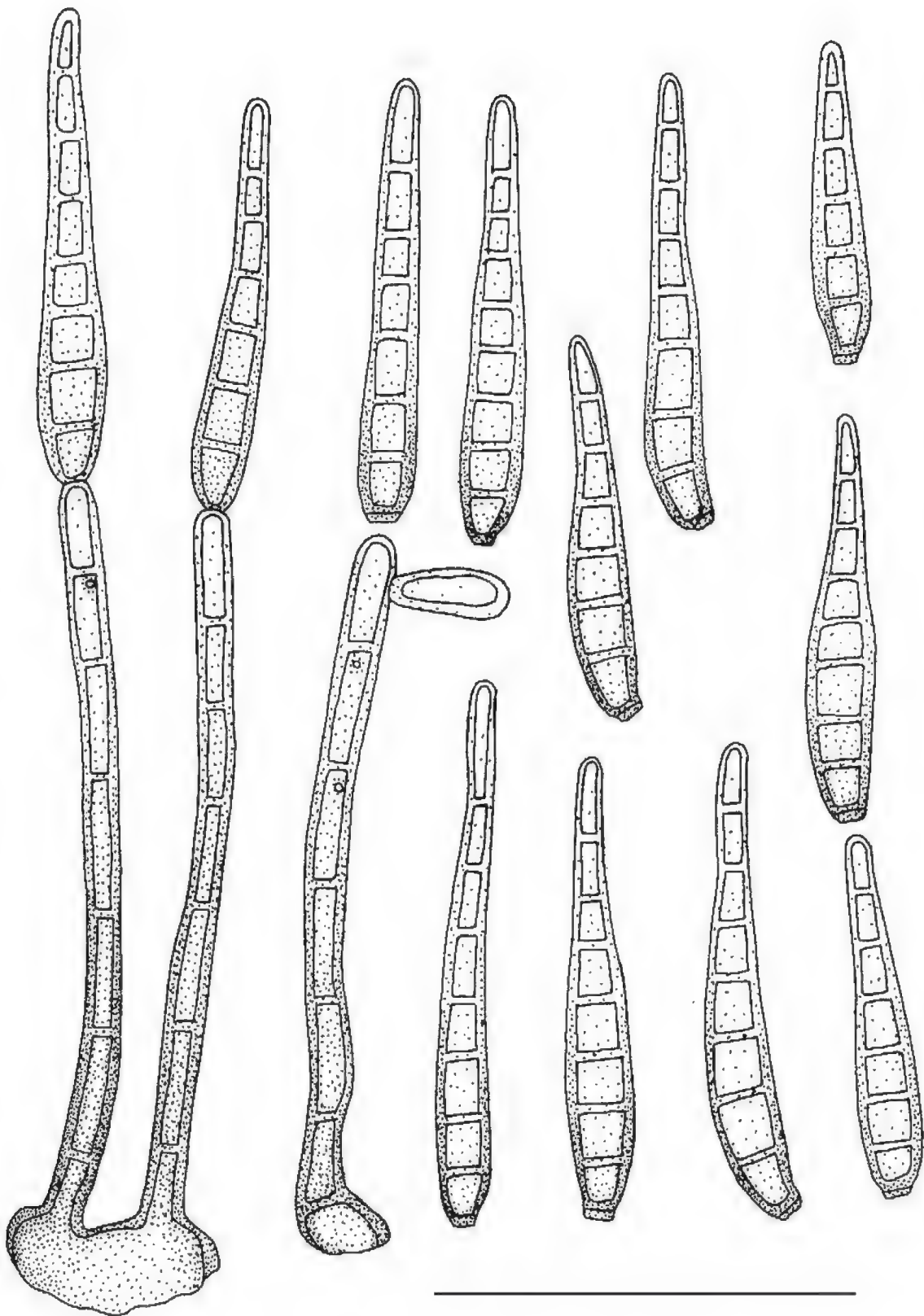


FIG. 2 *Helminthosporium hunanense* (ex holotype, bar = 50  $\mu$ m)  
Conidia and conidiophores on natural substratum

## Acknowledgements

The authors are grateful to Drs. R. F. Castañeda Ruiz, Instituto de Investigaciones Fundamentales en Agricultura Tropical “ Alejandro de Humboldt (INIFAT), Cuba and Prof. Y.L. Guo, Institute of Microbiology, Academia Sinica for reviewing the manuscript.

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## MYCOTAXON

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***Glomus candidum*, a new species of arbuscular mycorrhizal fungi from North American grassland**EDUARDO FURRAZOLA, RICARDO HERRERA-PERAZA<sup>§</sup>,*Instituto de Ecología y Sistemática, IES-CITMA**A.P. 8029, C. de La Habana 10800, Cuba*WITTAYA KAONONGBUA & JAMES D. BEVER<sup>\*</sup>*\*jbever@indiana.edu**Department of Biology, Indiana University**Bloomington, IN 47405, U.S.A.*

**Abstract** — A new species of arbuscular mycorrhizal fungi, *Glomus candidum* is described. The species produces spores singly in the soil. Spores are white to very pale yellow, usually globose to subglobose, 87–157 µm diam. Spore wall consists of two adherent layers. The outer layer is hyaline, mucilaginous, and stains very pale pink in Melzer's reagent. This layer can be observed in young spores and often degrades at maturity. The inner layer is hyaline and laminated, but occasionally the innermost group of laminae are pigmented a pale yellow to give the impression of two separated layers.

**Resumen** — Se describe una nueva especie de hongo formador de micorrizas arbusculares, *Glomus candidum*. La especie produce esporas libres en el suelo. Las esporas son blancas a amarillo muy pálido, usualmente globosas a subglobosas, 87–157 µm de diámetro. La pared de la espora consiste en dos capas adherentes. La capa externa a menudo se degrada con la madurez, es hialina, mucilaginosa, y se tiñe, sólo en las esporas jóvenes, de rosado muy pálido en reactivo de Melzer. La capa interna es hialina y laminada, pero a veces el grupo más interno de láminas aparece pigmentado de amarillo claro dando la impresión de dos capas separadas.

**Key words** — classification, molecular phylogeny, species description, taxonomy

**Introduction**

In studies of arbuscular mycorrhizal (AM) fungal ecology in an old field plant community on the campus of Duke University in Durham, North Carolina, a new species of *Glomus* was discovered with spores that were white to opaque

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<sup>§</sup> Deceased

when old (Bever et al. 1996, 2001). This fungus was subsequently used in multiple experiments on plant-soil feedback (Bever 2002), context dependence of plant growth promotion (Reynolds et al. 2005, Reynolds et al. 2006), effects on plant tolerance and defense against above-ground herbivores (Bennett & Bever 2007, Bennett et al. 2009), fungal competition (Bennett & Bever 2009) and preferential plant allocation (Bever et al. 2009). In the present paper, we describe this as a new species, *Glomus candidum*, sp. nov. based on morphology of mature spores as defined for the genus *Glomus* by Morton (1996) and Stürmer & Morton (1997), and the molecular studies of sequence from the large subunit (LSU) of the nuclear ribosomal (nrRNA) gene.

### Materials and methods

Samples consisting of soil and roots fragments were collected from the rhizosphere of *Allium vineale* L., *Anthoxanthum odoratum* L., *Plantago lanceolata* L. and *Panicum sphaerocarpon* Elliott growing in the field plot. Soils were mixed 1:1 (v/v) with silica sand that had been autoclaved for 1 hour at 120°C, then placed in greenhouse Deepots™ (Stuewe and Sons, Corvallis, OR), and seeded with either *Sorghum halepense* (L.) Pers. or *Sorghum bicolor* (L.) Moench separately. In addition, rhizosphere soil was collected to establish trap cultures using the above field plant species as hosts as described by Bever et al. (1996). Cultures were maintained in a cool season greenhouse (4–21°C) at Duke University. After 20 wk and once dried in situ, pot contents were harvested and stored at 4°C for 2 mo. Sporulation of the new *Glomus* sp. was particularly abundant with *Pa. sphaerocarpon* (note *Glomus candidum* = *Gl.* sp. D1 in Bever et al. 1996, = *Gl.* “white” in Bever et al. 2009, = *Gl. hyalinulum* (ined.) in Msiska & Morton 2009).

Spores were extracted from soil by wet sieving and decanting followed by centrifugation in a 20–60% sucrose density gradient (Daniels & Skipper 1982). Healthy spores were pipetted onto roots of 10–12 d old *S. bicolor* seedlings. Each inoculated seedlings were then transplanted into 4 × 21 cm Cone-tainers™ (Stuewe and Sons, Corvallis, OR) containing a sterile loamy soil:sand mix (1:2 v/v), adjusted to pH 6.2 and grown for 120 d in a growth room at the International Culture Collection of (Vesicular) Arbuscular Mycorrhizal Fungi (INVAM) at West Virginia University with a temperature range of 21–28°C, 225 μmol m<sup>-2</sup>s<sup>-1</sup> light intensity and a 14-h photoperiod.

Once monospecific cultures of *G. candidum* was successfully established, the contents of selected Cone-tainers™ were placed in the center of 15-cm pot, surrounded with the same growth medium described earlier, seeded with sudan grass (*Sorghum sudanense* (Piper) Stapf) and grown for another five months. This isolate (deposited in INVAM as NC268) and single spore cultures, maintained as pure cultures for more than ten years in the Bever lab collection, were used to describe the new species, *G. candidum*.

Spore size was measured with an ocular micrometer and color of spores was determined under reflected light from a two-branch fiber optic illuminator (color temp 3400 K) to co-illuminate spores and a printed color chart (INVAM color chart). The colors on the chart were composed of various percentages of the component colors: cyan, magenta, yellow and black. Spores were preserved in 0.05% NaNO<sub>3</sub> at 4°C.

Spores were also mounted in polyvinyl alcohol lacto-glycerol (PVLG) (Koske & Tessier 1983) and PVLG mixed with Melzer's reagent (1:1 v/v) to observe and measure spore subcellular structures. Slides of the spores were incubated in a conventional oven at 65°C for 24–48 hours and deposited as permanent vouchers at Oregon State University (OSC), Corvallis, OR; Harvard University (FH), Cambridge, MA; INVAM, the Bever lab collection and personal collection of W. Kaonongbua. Selected images of the spores were captured by a Sony CCD video camera on a Nikon Eclipse E600 Microscope. Color images in this paper are available from the corresponding author upon request.

The LSU nrRNA gene sequence (GU980757) was obtained from a single spore of the new fungus and was similarly subjected to various analyses (BLAST search, similarity index) as previously described (Kaonongbua et al. 2010). Additional LSU nrRNA sequences broadly representing the phylum Glomeromycota and outgroups (*Mortierella polycephala* – AF113464 and *Basidiobolus ranarum* – AF113452) were acquired from the NCBI's GenBank and then aligned using Clustal X (v. 2.0) (Larkin et al. 2007). After manual inspection and editing, a phylogenetic reconstruction based on the Neighbor-Joining (NJ) method with Kimura's 2-parameter model of nucleotide substitution was performed using MEGA4 (Tamura et al. 2007) with 1000 bootstrap replications.

## Taxonomy

***Glomus candidum*** Furrázola, Kaonongbua & Bever, sp. nov.

FIG. 1

MYCOBANK MB516798

*Sporocarpia ignota*. Sporae in solo singulatim efformatae, terminales, globosae vel subglobosae, 87–157 µm diam., candidus vel pallide luteae. Sporae tunica strata duabus: stratis exterior hyalino, immundo, caduco, 1.7–4.3 µm crasso; stratis secundo hyalinae vel pallide lutei, subtiliter laminato, 3.7–9.4 µm crasso. Hyphae subtendentes 6–21 µm diam., rectae vel recurvatae, porus septatus. Hyphae tunica strata duabus. Mycorrhizae vesicular-arbusculares formans.

**HOLOTYPE:** UNITED STATES. NORTH CAROLINA: Durham County on Duke University Campus, from an old field on the corner of Alexander Drive and University Avenue. September 1992, JD Bever, from culture IU-06. Deposited at OSC as broken spores mounted permanently on a glass slide, and labeled as 'holotype'.

**ETYMOLOGY:** from the Latin: '*candidus*' (white) referring to the white color of the spores under stereomicroscope.

Spores formed singly in soil and roots; globose to subglobose, 87–157 µm diam (mean = 125 µm, n = 125); white, with a few spores becoming a pale yellow color (0-0-5-0) with age. Spores have a thin "halo" under reflected light (FIG. 1.1) when all spore wall layers are present. The spore wall consists of two adherent layers (FIGS. 1.2–1.3). The outer layer (L1) is hyaline, mucilaginous initially, becoming more granular as it begins to decompose, 1.7–4.3 µm thick, often adherent to the inner structural laminate layer, staining very pale pink in Melzer's reagent in juvenile spores only. With age, this layer degrades and decomposes naturally, after which it appears granular and may accumulate some debris. The inner layer (L2) is laminated, 3.7–9.4 µm thick (mean = 6.1,



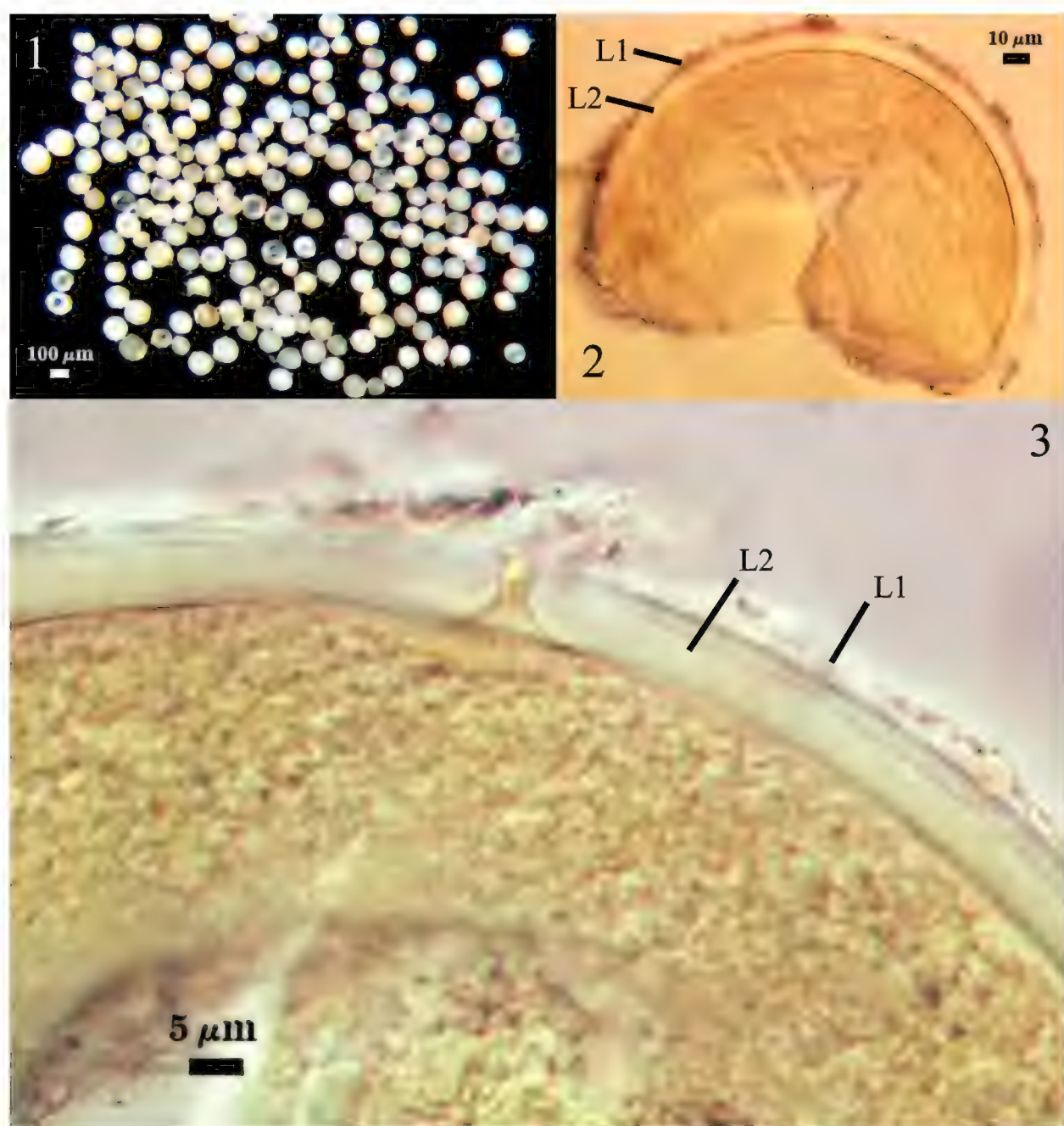


FIGURE 1. *Glomus candidum*, Reference Accession NC268. 1, Color and shape of spores under the dissecting microscope; note the halo around some spores. 2, Spore wall composed by two layers (L1 and L2). 3, A larger magnification showing an improved view of wall composition.

n = 40). It is often uniformly hyaline, but in a few spores, an innermost group of laminae (sublayers) is pigmented a pale yellow (0-15-0-0) to give the appearance of two separates layers of occasionally equal thickness. This color separation is not consistent in all spores. No part of the laminate layer reacted in Melzer's reagent. Subtending hyphae is single, straight or occasionally recurved, cylindrical to slightly flared, 6–21 μm wide at spore base. Some spores lack the subtending hyphae due to breakage close to the spore base. In mature spores, the innermost sublayer(s) of the laminate layer of the spore wall, usually forms a thin septum (1.0–1.7 μm thick); positioned 2–16 μm in the hyphal lumen. In

some spores, the occlusion is a hyaline plug, or the lumen of subtending hyphae remains open. The subtending hyphal wall consists of a continuation of both layers of the spore wall. The L2 layer tapers gradually to 1  $\mu\text{m}$  thick, 20–25  $\mu\text{m}$  from the spore base.

MYCORRHIZAE: *G. candidum* has been observed to form arbuscules and vesicles typical of *Glomeraceae*.

DISTRIBUTION AND HABITAT: This species is known from an old field on the campus of Duke University, Durham County, North Carolina. Soil pH in this field averaged 5.2. Soil phosphorus ranged from 7.6 to 48.9  $\text{kg}\cdot\text{ha}^{-1}$  and averaged 17.8  $\text{kg}\cdot\text{ha}^{-1}$ . The average percent organic matter at the sampling sites was 4.2 and the range varied from 1.07 to 14.13. Fungi with similar morphology have also been isolated from Indiana, Maryland, and West Virginia.

MYCORRHIZAL ASSOCIATIONS: Found in the rhizosphere of *Al. vineale*, *An. odoratum*, *Pl. lanceolata*, and *Pa. sphaerocarpon* in the field plot. Formed arbuscular mycorrhizae on *S. bicolor*, *S. halepense*, *S. sudanense*, and *Zea mays* in greenhouse-grown pot cultures.

MOLECULAR PHYLOGENETIC ANALYSIS: The LSU nrRNA gene tree clearly places the new fungus as a member of the *Glomus* group B (Schwarzott et al. 2001) along with sequences from *Glomus claroideum* N.C. Schenck & G.S. Sm., *Glomus etunicatum* W.N. Becker & Gerd., and *Glomus luteum* L.J. Kenn. et al. and with 100% statistical support (FIG. 2).

## Discussion

Our phylogenetic analysis of the LSU nrRNA gene sequences placed *G. candidum* in *Glomus* Group B sensu Schwarzott et al. (2001). This is consistent with the phylogeny generated by the  $\beta$ -tubulin gene using an isolate of *G. candidum* from Maryland (Msiska & Morton 2009). Both of these studies were limited by using only a single sequence from a single spore of the new AM fungal species. Thus the molecular work cannot test the extent of genetic divergence within this species or between *G. candidum* and other species in this clade.

Spores of *G. candidum* differ in morphology from other species in *Glomus* Group B, including *G. claroideum*, *G. lamellosum* Dalpé et al., and *G. luteum*. Spore size range of *G. candidum* overlaps with that of *G. claroideum* (87–157 and 70–180  $\mu\text{m}$ , respectively); however, both species differ in their spore ontogenies. Whereas fully formed spores of *G. claroideum* show four layers (Stürmer & Morton 1997), spores of *G. candidum* develop only two layers. Occasionally, the innermost group of laminae (sublayers) of *G. candidum* spore wall layer 2 may appear as a separate layer due to slight pigmentation. In addition, the pore of the subtending hypha of *G. claroideum* spores is occluded either by a sublayer (lamina) of the laminate spore wall layer 3 and spore wall

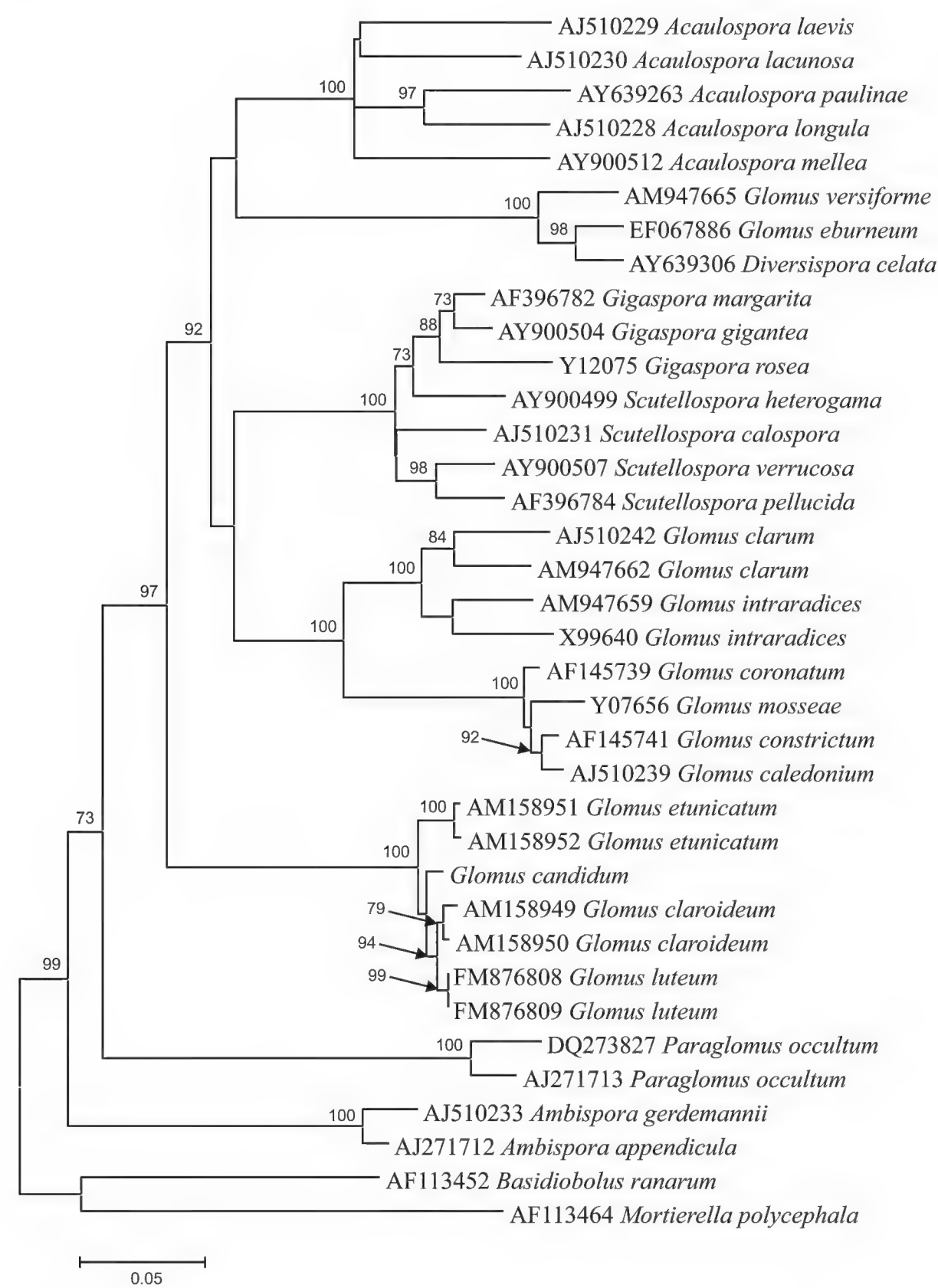


FIGURE 2. Neighbor-joining tree inferred from partial nrLSU sequences showing the taxonomic position of *Glomus candidum* as a member of the *Glomus* “Group B”. The numbers on the tree braches are bootstrap support values based on 1000 replications of the neighbor-joining analysis (values are not shown if it is below 70%). Sequences of *Mortierella polycephala* (AF113464) and *Basidiobolus ranarum* (AF113452) were used as outgroups. The scale indicates the number of base substitutions per site.

layer 4 or only by spore wall layer 4. In contrast, in *G. candidum* the closure is by the presence of a thin septum or a mucilaginous plug.

When viewed under a dissecting microscope, *G. candidum* spores also resemble those of *G. lamellosum* because of their similar size range (87–157 and 98–142 × 122–162 µm, respectively) and their whitish to pale yellow pigmentation (Dalpé et al. 1992). However, *G. lamellosum* spores have been described with a spore wall composed of three layers persisting in mature spores, with the outermost one significantly thicker than the first layer of *G. candidum* (4–14 µm and 1.7–4.3 µm thick, respectively) and the flexible innermost layer not observed in the species being described here.

*Glomus luteum* forms spores slightly bigger than those formed by *G. candidum* (up to 180 µm in diam) and *G. luteum* spores have been described with four layers, including a flexible innermost layer (Kennedy et al. 1999), which is lacking in the spore wall of *G. candidum*. In addition, *G. luteum* spores have been described as pale yellow to dark yellow with a brownish tint in color compared to the white spores of *G. candidum*. With age, spores of *G. candidum* may turn pale yellow, but examinations of crushed spores under a compound microscope can readily separate *G. candidum* from the three species listed above based on the number of layers of their spore wall. In addition, in PVLG + Melzer's, the outermost layer of *G. candidum* reacts very pale pink while the reactions are darker pink in *G. claroideum*, pinkish-red in *G. luteum* and non-reactive in *G. lamellosum*.

Spores of *G. candidum* bear superficial similarity to other phylogenetically unrelated *Glomus* species producing white/hyaline spores, including *G. diaphanum* J.B. Morton & C. Walker, *G. clarum* T.H. Nicolson & N.C. Schenck, and *G. manihotis* R.H. Howeler et al. under a stereomicroscope. The spore wall structure of *G. candidum* was initially judged most similar to *G. diaphanum* according to Bever et al. (1996). However, even though spore wall layers 1 and 2 of both species are similar in their phenotypic and biochemical properties, the latter species also has a flexible layer 3 in the spore wall, which is missing in the former species.

On the rare occasions in which the innermost group of laminae (sublayers) of spore wall layer 2 of *G. candidum* is pigmented, its spore can resemble those of *G. clarum* and *G. manihotis* (Stürmer & Morton 1997). This can be particularly difficult to discern under low magnification, but *G. candidum* spores never reach as dark a tint as do those of *G. clarum* and *G. manihotis*. Spores of *G. clarum* and *G. manihotis* are also bigger (up to 260 µm in diam) than those of *G. candidum*, whose biggest spores reach approximately 160 µm in diam. In addition, in *G. clarum* and *G. manihotis* the external mucilaginous layer reacts strongly in Melzer's reagent (pinkish-red to light purple), while the reaction of the same layer in *G. candidum* is very pale pink and only occurs in juvenile



spores. Finally, molecular tools clearly identify that *G. candidum* is a member of *Glomus* group B, while *G. diaphanum*, *G. clarum*, and *G. manihotis* are in *Glomus* group A sensu Schwarzott et al. (2001) (Msiska & Morton 2009).

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## MYCOTAXON

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**New record of *Circinella muscae* from a hydrocarbon polluted sand beach of Tabasco, Mexico**MARÍA C. GONZÁLEZ<sup>1\*</sup>, NAYELI MURUETA-FIGUEROA<sup>1</sup>,  
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**Abstract** — During a survey of fungal biodiversity from Mexican sand beaches, an uncommon fungus of the subphylum *Mucoromycotina* was isolated from the intertidal area of Playa Paraiso, State of Tabasco. A study of culture isolates demonstrated that it is a mucoraceous species belonging to the genus *Circinella* characterized by sporangiophores bearing circinate branches terminated by globose sporangia with persistent sporangial walls. Several sandy soil samples placed in sterile re-sealable plastic bags were processed in the laboratory within 4 h. Plates of corn meal agar inoculated with 0.5 g of sandy soil were incubated 15 d. The fungus produced sympodially branched sporangiophores with fertile circinate branches bearing one or two sporangia, or a single sporangium and a sterile spine. Sterile spines were light in color and the globose sporangia had persistent walls bearing globose, hyaline sporangiospores. The characters of the Mexican isolate agree with those described for *C. muscae*. Few zygomycete studies have been conducted in Mexico, making this the first recorded mucoraceous fungus isolated from a sand beach environment in the country.

**Key words** — arenicolous fungi, endopsammon, Gulf Coast of Mexico, tropical seashore

**Introduction**

At the national level, little is known about fungal communities that inhabit the endopsammon in Mexico (González et al. 1998, 2000). During a survey of fungal biodiversity from Mexican sand beaches, an uncommon fungus of the phylum *Zygomycota* was isolated from the intertidal area of Playa Paraiso, State of Tabasco. A study of the characteristics of this isolate on culture media demonstrated that it is a mucoraceous species belonging to the genus *Circinella*



Tiegh. & G. Le Monn., characterized by the production of sporangiophores bearing circinate branches terminated by globose multispored sporangia with persistent sporangial walls. After the genus was monographed by Hesseltine & Fennell (1955), several additional species were described (Hesseltine & Ellis 1961, Faurel & Schotter 1965, Patil & Kale 1981, Arambarri & Cabello 1996). *Circinella* currently includes nine species with an apparently worldwide distribution (Benny 2006). Members of this genus usually have been isolated from soil, dung, fermented cacao beans, musty nuts, and more recently, *C. lacrymispora* was described from hydrocarbon-polluted soil from the coast of Argentina (Arambarri & Cabello 1996). The biotechnological potential of *Circinella* has been explored by Nakagawa et al. (1995) who performed the biochemical conversion of milbemycins with *C. umbellata*.

### Materials & methods

Playa Paraiso is located on the southeast coast of the Gulf of Mexico (18°24'00"N 93°13'59"W) in the County of Paraiso, a petroleum extraction zone characterized by numerous lagoons, estuaries, and swamps. It receives 1,751 mm of annual precipitation with a median annual temperature of 26°C. In this study, the beach was sampled August 18, 2001, during low tide; three sandy soil samples of 50 g each were collected and placed in sterile Ziploc® bags and were processed in the laboratory within 4 h. The surface of six plates of corn meal agar (Difco) prepared with artificial seawater (Instant Ocean®) with antibiotics (chloramphenicol 1 mg/ml, penicillin 500 µ/ml, streptomycin 300 µg/ml added after autoclaving) were inoculated each one with 0.5 g of sandy soil and incubated 15 d at 25°C. After this incubation, a mucoraceous fungus was transferred to mucor agar (Hesseltine 1954, Hesseltine & Fennell 1955), potato dextrose agar, malt agar and V8 vegetable juice agar without antibiotics for descriptive purposes. The fungus was identified with the keys published by Hesseltine & Fennell (1955) and Arambarri & Cabello (1996). The culture and slide of this isolate are deposited in the fungal collection of Herbario Nacional (MEXU) of the Institute of Biology, Universidad Nacional Autónoma de México.

### Results

A total of 28 fungal isolates were recovered from the sandy soil samples of Paraiso Beach, and 42 colony-forming units were obtained per gram of soil. *Circinella muscae* was an uncommon species with a low relative abundance value (0.14%).

*Circinella muscae* (Sorokīn) Berl. & De Toni, Sylloge Fungorum 7: 216. 1888.

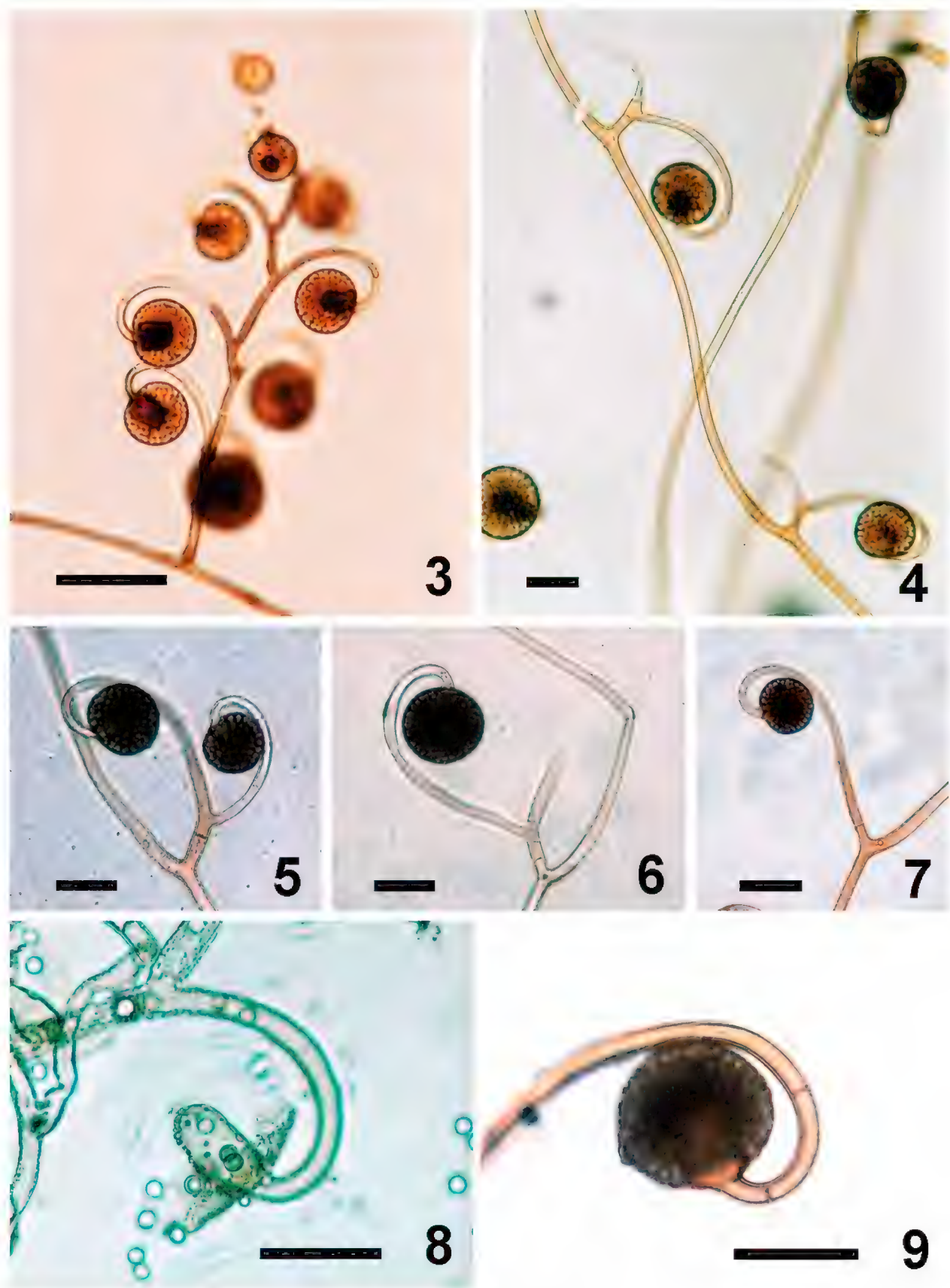
FIGS 1–9

SPECIMEN EXAMINED: MEXICO. State of Tabasco: Paraiso Co., Paraiso Beach (18°24'00"N 93°13'59"W), from intertidal soil sand, 18 Aug 2001, *N. Murueta-Figueroa*, *M.C. González*. MEXU 25512, dehydrated specimen, slide and culture MGSW20.



FIGS. 1, 2. *Circinella muscae*. 1. Colony appearance on V8 vegetable juice agar after 7 days at 25°C. 2. Colony appearance on potato dextrose agar after 7 days at 25°C. Petri dishes: 100 mm diam.

Mycelium forming a uniform, dense colony on the surface of the agar, relatively slow-growing, reaching a diam. of 75 mm after 7 days at 22–23°C. Colony



FIGS. 3–7 *Circinella muscae*. 3. Lateral branch of aerial sporangiophore with a sympodial arrangement of small sporangia. 4. Aerial sporangiophore showing lateral branches with sterile spines and circinate sporangia. 5–7. Aerial sporangiophore with lateral branches bearing two sporangia, a sporangium and a sterile spine, or one sporangium. 8. Columella of sporangium with basal collar and sporangiospores. 9. Characteristic two septa near end of lateral sporangial branch. All bars = 50 µm.



initially white, then gray, becoming cinnamon brown with age (FIGS 1, 2). Colony consisting of a layer of short sporangiophores overgrown by longer, aerial sporangiophores. Short sporangiophores erect, up to 1 cm high, arising from hyphae on the agar surface, nonseptate, recurving at apex to form a terminal sporangium. Short sporangiophores with closely spaced, sympodial branches, each with an apical circinate sporangium, frequently with a few large sporangia and a cluster of smaller sporangia. Aerial sporangiophores very long, up to 6 cm in length, sometimes with two main branches, straight to wavy, but never spirally coiled. Aerial sporangiophores with scattered, alternate lateral branches terminated by a large circinately attached sporangium, or dichotomously branched, with two sporangia, a sporangium and a sterile spine, or occasionally with two sterile spines; a few lateral branches bear a sympodial arrangement of small sporangia and without spines (FIGS 3–7). Sporangial branches usually with a basal septum and sometimes with one or two additional septa near the sporangium (FIG. 9). Sporangia globose, nonapophysate, with a columella and persistent wall, at first hyaline, then dark gray and appearing black in reflected light. Large sporangia 46–72(–92)  $\mu\text{m}$  diam (sd = 12.9, n = 25), small sporangia 18–28  $\mu\text{m}$  diam (sd = 4.9, n = 25), the different sizes readily discernable under the dissecting microscope. Sporangiophores and sporangial branches light brown in color. Columellae variable in shape, subglobose, oblong or conical, smooth, sometimes with a short, hyaline apical protuberance, 21–42  $\mu\text{m}$  high  $\times$  15–32  $\mu\text{m}$  wide. A large, broad collar often remains around the base of the columella after breakdown of the sporangial wall (FIG. 8). Sporangia filled with numerous sporangiospores that are liberated when the wall eventually breaks up. Sporangiospores globose, subglobose to ovoidal, 5–7  $\times$  5–6.6  $\mu\text{m}$ , hyaline and smooth, appearing dark brown to black in mass. Zygosporangia not observed.

## Discussion

The tropical species *Circinella mucoroides* Saito is morphologically close to *C. muscae* in producing subglobose, multisporous sporangia on circinate sporangiophores, globose sporangiospores, and the presence of sterile spines. However, *C. mucoroides* has spirally twisted sporangiophores that often bear branches that only form spines, whereas *C. muscae* has straight to wavy sporangiophores with branches that typically bear sporangia along with sterile spines. The change in colony color from gray to brown with age also is characteristic of *C. muscae*.

Although no extensive studies of zygomycetes have been conducted in Mexico, several other species of zygomycetes have been reported from the country. Benny & Benjamin (1975, 1976) isolated *Backusella ctenidia* (Durrell & M. Fleming) Pidopl. & Milko ex Benny & R.K. Benj., *Benjaminiella poitrasii*



(R.K. Benj.) Arx (as *Mycotypha poitrasii* (R.K. Benj.) Benny & R.K. Benj.), *Chaetocladium brefeldii* Tiegh. & G. Le Monn., *Cokeromyces recurvatus* Poitras, *Dichotomocladium elegans* Benny & R.K. Benj., *D. robustum* Benny & R.K. Benj., *Thamnostylum lucknowense* (J.N. Rai et al.) Arx & H.P. Upadhyay, *T. nigricans* (Tiegh.) Benny & R.K. Benj. and *Zychaea mexicana* Benny & R.K. Benj. from rodent and lizard dung in northern Mexico. Earlier, Zenteno-Zevada et al. (1955) reported *Rhizopus* sp. on *Annona* sp. in Veracruz and *R. stolonifer* (Ehrenb.) Vuill. (as *R. nigricans* Ehrenb.) on potato from Chihuahua and Guanajuato. Ulloa & Herrera (1971) isolated *R. stolonifer* and *Mucor racemosus* Fresen. from pozol; and Pérez-Silva (1976) isolated *Thamnostylum piriforme* (Bainier) Arx & H.P. Upadhyay (as *Helicostylum piriforme* Bainier) from cow dung from the Distrito Federal. Samaniego et al. (1988) isolated *Actinomucor elegans* (Eidam) C.R. Benj. & Hesselt., *Mucor* sp., *Phycomyces* sp., and *Rhizopus arrhizus* A. Fisch. from soils in Coahuila State. Moretti & Robledo (1988) isolated *Mucor* sp., *Rhizopus* sp., and *Syncephalastrum* sp. from air samples in Mexico City in the Distrito Federal; and Ramírez-Guillen & Guzmán (2003) reported *Thamnidium elegans* Link on a decaying basidioma of *Lepiota* sp. from Veracruz. Trigos et al (2008) isolated *Circinella minor* Lendn. from “ejote” (*Phaseolus vulgaris* L.), also in Veracruz.

The physical and chemical properties of a soil may determine the fungal diversity that inhabits that ecosystem. The fungal diversity of coastal sand beaches still is unknown for the most part. The sandy soil of beaches probably has a high and characteristic mycobiota composed of species adapted to that particular marine environment where the ascomycetes are the more common and best studied group (Kohlmeyer & Kohlmeyer 1979, Dunn & Baker 1983). This is the first record of a mucoraceous fungus isolated from a sand beach environment in Mexico. Because this fungus was isolated from hydrocarbon polluted sand, chemical studies need to be performed to investigate its potential biotechnological value.

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## MYCOTAXON

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**Two *Oudemansiella* species with echinulate basidiospores from South America with *O. macracantha* lectotypified**

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**Abstract**—A recent collection of *Oudemansiella steffenii* from the State of Pernambuco, Brazil, is described and compared to the type of *O. macracantha*, which is lectotypified here. Photographs of both basidiomes and microstructures are also provided.

**Key words**— *Agaricales*, Neotropics, *Physalacriaceae*, taxonomy

## Introduction

Recent molecular and phylogenetic studies showed that species covered by dense long hairs, such as *Xerula hispida* Halling & G.M. Muell. (1999) and *X. setulosa* (Murrill) R.H. Petersen & T.J. Baroni (2007), form a well supported clade while non-hispid species of *Xerula* Maire and *Oudemansiella* Speg. s. str. taxa form another (Mueller et al. 2001). The bootstrap value between these clades is low and the hairy *Xerula* and all other *Oudemansiella* sensu Singer (1986) represent two distinct genera also supported by recent morphological studies (Wang et al. 2008, Yang et al. 2009).

Among the sections proposed in the rearrangement of *Oudemansiella* by Yang et al. (2009), the species with echinulate basidiospores, *O. steffenii* and *O. macracantha*, initially placed at the subgenus level by Cléménçon (1979), belong to sect. *Dactylosporina* (Cléménçon) Pegler & T.W.K Young, as already reported by Pegler & Young (1986).



In this study, recent collections of *O. steffenii* and the type of *O. macracantha* were studied in order to clarify the species concepts among the taxa of sect. *Dactylosporina*.

### Materials and methods

Microscopic observations were made from material mounted in 3% KOH and Congo Red solutions. Presentation of basidiospore data follows the methodology proposed by Tulloss et al. (1992), but utilizing a single basidioma (Wartchow 2009). The notation “[*a/b/c*]” at the beginning of the spore data set is to be read “*a* spores measured from *b* basidiomes taken from *c* collections.” Other abbreviations include L(W) = basidiospore length (width) average from a single basidiome, Q = the length : width ratio range as determined from all measured basidiospores, and  $\bar{Q}$  = the Q value averaged from all basidiospores measured within a single basidiome. Color codes used in the description of the species are those from Watling (1969). Herbaria codes and names follow Holmgren & Holmgren (2001). Description of dried material of *O. macracantha* follows the style of the type studies of Yang (2000).

### Taxonomy

*Oudemansiella steffenii* (Rick) Singer, Lilloa 26: 66. 1953.

FIGS. 1–7

MATERIAL EXAMINED: BRAZIL. Pernambuco, Recife, Campus UFPE, 01.vi.2007, *J. Pereira s.n.* (URM 79226).

Basidiome medium-sized. PILEUS to 65 mm in diam., plano-umbonate to concave-umbonate, brown (‘milk coffee’ 26) to slightly paler (‘snuff brown’ 17) at margin, narrowly sulcate-striate (5–10 mm) when fresh, more indistinct in dried state; context thin, fleshy. LAMELLAE adnexed to somewhat sub-free, white to cream, somewhat brown at edges, 4–8 mm wide, subdistant; lamellulae very common, 14–20 mm long, frequently truncate. STIPE 95 × 4–8 mm, cylindrical above bulb to tapering near apex, brown (‘clay buff’ 32 to ‘snuff brown’ 17); bulb inflating to 6–12 mm, pseudorrhiza very long, 25–100 mm; context pale cream, solid.

BASIDIOSPORES [25/1/1] 12–15.5 × 11–14.5 µm (without ornamentation), L = 13.2 µm, W = 12.7 µm, Q = 1.00–1.11(–1.16),  $\bar{Q}$  = 1.04, globose only infrequently subglobose, moderately thick walled, strongly spinose with > 30 spines (2–)3.5–5(–5.5) µm long, subacute to subobtuse, infrequently with acute tips, inamyloid, colorless, with guttulate contents. BASIDIA 50–60 × 13–15 µm, clavate, 4-sterigmate, sterigmata to 9 × 4.5 µm (width measured at base). PLEUROCYSTIDIA scattered 60–120 × 18–36 µm, fusoid to lageniform, rounded-obtuse to subcapitate, infrequently subacute, thick-walled (1.5–3 µm), hyaline, colorless. CHEILOCYSTIDIA not observed. PILEIPELLIS a hymenoderm layer consisting of elements 22–45 × 14.5–22 µm, somewhat to broadly clavate or more or less pyriform (e.g. 30 × 11 µm or 70 × 22 µm), all rounded-obtuse at

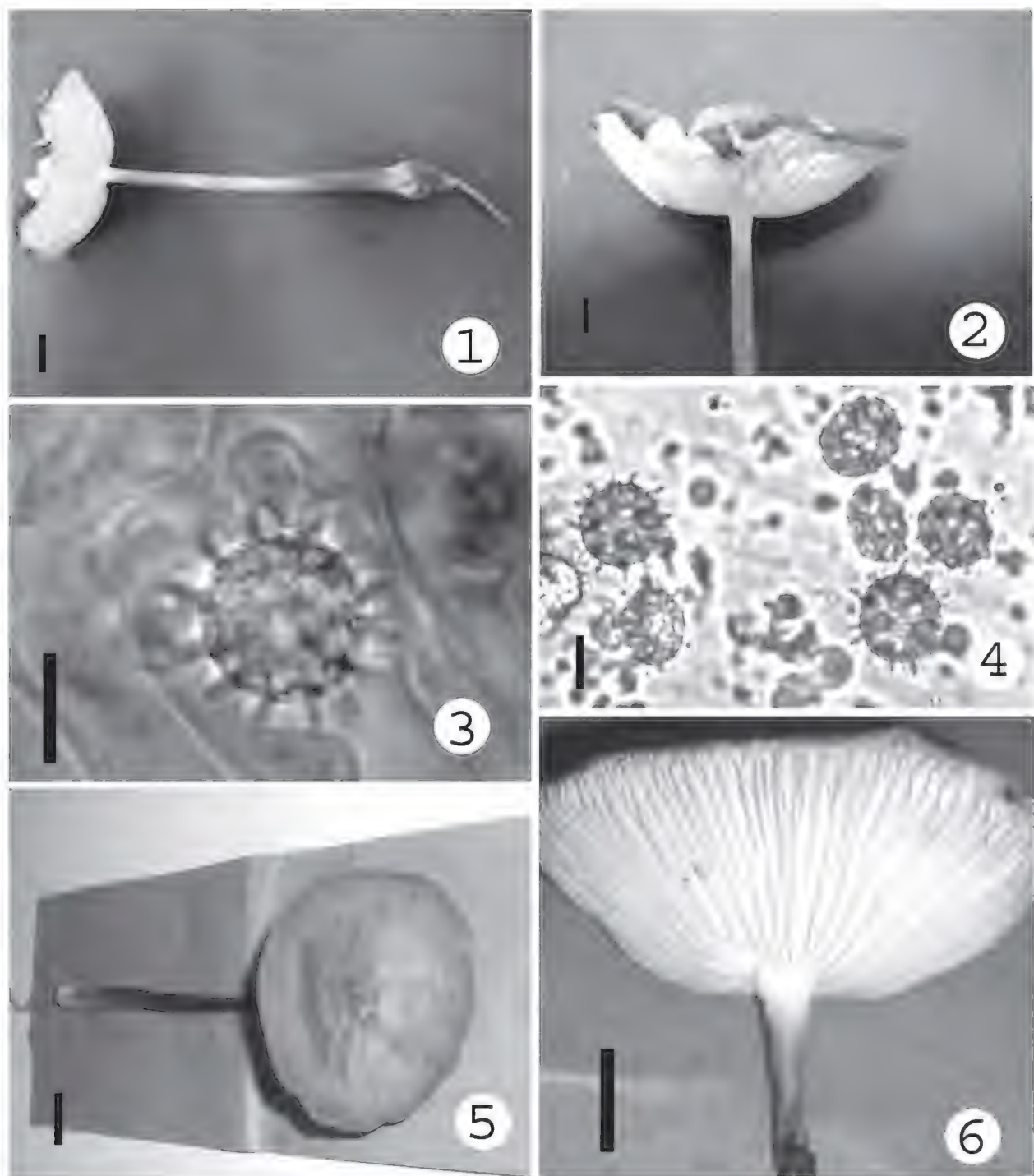


FIG. 1–6. *Oudemansiella steffenii*. 1–2. Basidiome (URM 79226). 3–4. Basidiospores (URM 79226). 5–6. Basidiome (URM 79227). Scale bar is 10 mm for basidiomes and 10  $\mu$ m for microstructures. (Photos: 1–4 by J. Pereira; 5–6 by A.C. Gomes-Silva).

apex, brownish or slightly paler pigmented, walls to 1  $\mu$ m thick, arising from a hypoderm of filamentous hyphae 4–8  $\mu$ m wide. LAMELLA TRAMA regular to somewhat sub-irregular, with filamentous hyphae frequently septate, 3–6  $\mu$ m wide, somewhat inflated to 13–18  $\mu$ m, occasionally clamped.

HABITAT: Solitary or scattered, on soil (attached to buried rotting wood?) in urban area.

ADDITIONAL SPECIMENS EXAMINED: BRAZIL, **Pernambuco**, Recife, 'Reserva Ecológica Dois Irmãos', 03.vii.2006, *F. Wartchow 10/2006* (URM 80090); São Vicente Férrer, 'Mata do Estado', 26.vi.2008, *V.R.M. Coimbra & F. Wartchow s.n.* (URM 80091); **Rio Grande do Sul**, Santa Maria, Camobi, 'Morro do Elefante', 20.i.2001, *F. Wartchow 05/01* (SMDB 9189); Vera Cruz, Travessa Dona Josefa, 26.v.1987, *J. Putzke s.n.* (HCB 12143); **Rondônia**, Porto Velho, PVH, 22.i.2008, *A.C. Gomes-Silva 348* (URM 79227).

DISTRIBUTION: Argentina, Bolivia, Costa Rica, Colombia, Mexico, Panamá, and Venezuela [Singer 1964, Halling & Mueller 1999 as *Xerula steffenii* (Rick) Boekhout & Bas, probably Petersen 2008 as *Xerula macracantha* (Singer) Boekhout & Bas]. Brazil: Amazonas, Minas Gerais, Paraná, Pernambuco, Rio Grande do Sul, São Paulo [Singer 1964, Putzke & Pereira 1988, Souza & Aguiar 2004, Capelari & Gugliotta 2005 as *Dactylosporina steffenii* (Rick) Dörfelt, Sobestiansky 2005, de Meijer 2006 as *Xerula steffenii*, Drechsler-Santos et al. 2007, Rosa & Capelari 2009 as *Dactylosporina steffenii*]. *Oudemansiella steffenii* is a new record from the State of Rondônia, North Brazil.

REMARKS: Our description of *O. steffenii* is based on material recently collected in Pernambuco. It differs from *O. macracantha* in the following: (1) the larger basidiomata, (2) a more robust stipe, and (3) more numerous spines in the basidiospores. On the other hand, our material of *O. steffenii* shares with *O. macracantha* the pileus color and relatively long spines in some of the basidiospores. For a better understanding of species concepts among the echinulate spored species of *Oudemansiella*, the type of *O. macracantha* from Bolivian Amazon region was analyzed.

Other materials of *O. steffenii* analyzed (HCB 12143, SMDB 9189, URM 79227) show some differences in comparison with the Pernambuco collection. The majority of basidiospores of these materials have spines that are only rarely longer than 3 µm, except for URM 80091, in which at least one basidiospore showed spines up to 5 µm long. Previous descriptions (Singer 1964, Putzke & Pereira 1988, Halling & Mueller 1999, Capelari & Gugliotta 2005) also describe shorter basidiospore spines for *O. steffenii* compared to *O. macracantha*.

In URM 79226 (described above) thicker walled cystidia were also observed compared to those observed by the authors cited above; wall thickness, however, is not taxonomically diagnostic, since other collections (e.g., URM 80091) also had thick-walled cystidia.

In URM 79226 and SMDB 9189, an entirely brown pileus was observed, and in URM 79227 (recently collected in Amazon Forest, state of Rondônia), grayish tints were present in the sulcate margin, and spines mostly 2–3 µm long (occasionally ranging to 3.5 µm long) were observed. The pileus color pattern and spine length are also obvious features of *O. steffenii*.

***Oudemansiella macracantha*** Singer, Sydowia 15: 59. 1962 ('1961'). FIGS. 8–12

MATERIAL EXAMINED: BOLIVIA, Vaca Diez, Depto. Beni, Guayaramerín, 16.iii.1956, *R. Singer B 1997* (BAFC 51670 lectotypus hic designatus); same place, 17.iii.1956, *R. Singer B 2112* (LIL).

PILEUS 6–8 mm in diam., plane, surface brown ('snuff brown 17') to vinaceous brown ('umber 18'), somewhat paler at center ('milk coffee 28'), margin entire. LAMELLAE adnate, subclose to subdistant, buff ('buff 52'), edge slightly darker; lamellulae rare or absent. STIPE to 80 × 0.5 mm (fragmented), cylindric but slightly tapering upward.

BASIDIOSPORES [40/2/2] (10–)11–15(–16) × (9.5–)10.5–14(–15) µm (without ornamentation), L = 13.4 µm, W = 12.8 µm, Q = 1.00–1.07(–1.17), Q = 1.04, globose, only infrequently subglobose or broadly ellipsoid, moderately thick walled, strongly spinose having about 23 spines mostly 4.5–5.5 µm, only occasionally 2–2.5 µm long and only occasionally to 7 µm long (in R. Singer B 2112), tips subacute, inamyloid, colorless, with guttulate contents. BASIDIA 67 × 22 µm, clavate, 4-sterigmate, up to 9 × 4.5 µm (width measured at base). PLEUROCYSTIDIA difficult to locate (probably due to age of material), 60–72 × 27 µm, fusoid, rounded-obtuse, wall slightly thickened, hyaline, colorless. CHEILOCYSTIDIA not observed. PILEIPELLIS a hymenoderm consisting of elements 22 × 17 µm, broadly clavate, occasionally narrowly clavate or more or less pyriform, all rounded-obtuse at apex, brownish pigmented. STIPITPELLIS covered by caulocystidia 27–95(–180) × 14.5–22.5(–27), common, fusoid-lageniform, brownish pigmented that is somewhat condensed. LAMELLA TRAMA regular or appearing somewhat subregular, with filamentous hyphae frequently septate, 2.5–5.5 µm wide, occasionally clamped.

HABITAT: On buried wood in tropical rain forests, rather common, but scattered, fruiting in rainy seasons (Singer 1964).

DISTRIBUTION: This species is restricted to the frontier Amazon region between Brazil and Bolivia.

REMARKS: *Oudemansiella macracantha* previously was known only from the Bolivian Amazon region (Singer 1964), although recently it was reported from Argentina and Mexico by Petersen (2008 as *Xerula macracantha*), who reported an additional feature that could segregate the echinulate spored *Oudemansiella*: in *O. macracantha*, the spines remain turgid in spite of the vacuum applied by electron microscope, while in *O. steffenii* they are partially collapsed after SEM preparation. This observation was entirely based on recently collected material and not the type. Petersen (2008) also reported that the number and length of basidiospore spines in *O. macracantha* were more numerous and longer than those of *O. steffenii*. This conclusion, however, does not match satisfactorily with Singer (1964) who cited 38–42 spines per basidiospore in *O. steffenii* and only 23 in *O. macracantha*. Fewer spines were also observed on the type. The images provided by Petersen (2008) probably correspond to *O. steffenii* due the relatively shorter basidiospore spines depicted compared to the type specimen.



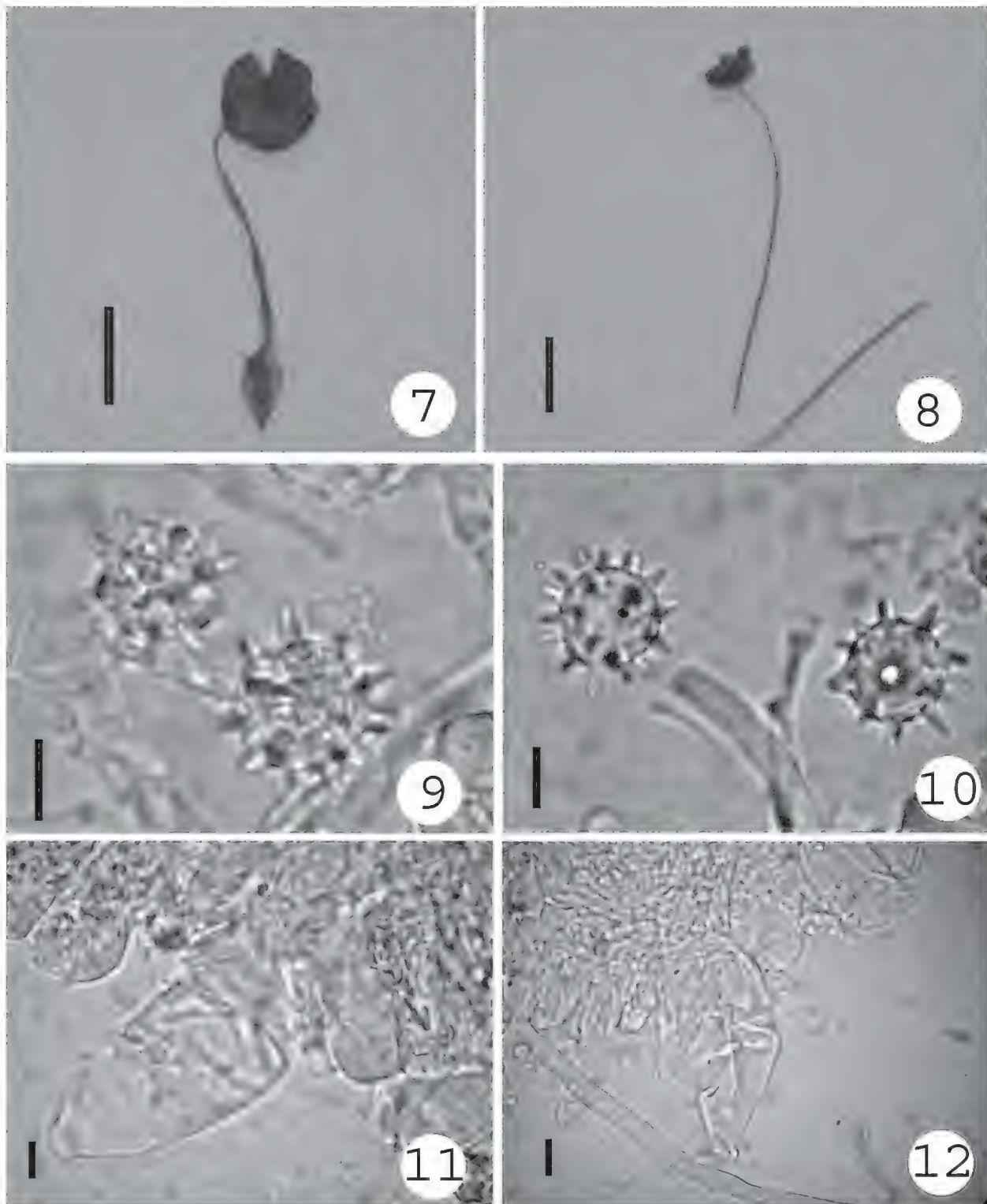


FIG. 7. *Oudemansiella steffenii*. Dry basidioma (HCB 12143).

FIG. 8–12. *Oudemansiella macracantha*.

8. Lectotype. 9–10. Basidiospores. 11–12. Pleurocystidia.

Scale bar is 10 mm for basidiomes and 10  $\mu$ m for microstructures.

(Photos: 7–12 by F. Wartchow).

The material from the State of São Paulo referred to as *O. macracantha* by Pegler (1997) is thought to represent *O. steffenii* by Capelari & Gugliotta (2005), although they did not study the type; the material from Paraná is missing (de Meijer 2006).

Summarizing, *O. macracantha* is a well defined species that Singer (1964) differentiated by the small pileus (< 15 mm in diam.), longer and much more slender stipe, and basidiospores with longer, less numerous spines.

*Oudemansiella macracantha* appears, in fact, restricted to its type locality. Some collections of *O. steffenii* (e.g., HCB 12143, URM 80090) also present a relatively slender basidiome like the type material of *O. macracantha*, but they have the proportionally shorter stipe and obviously shorter spines that are more commonly to *O. steffenii*.

We also address in this paper the issue of the type collection for *O. macracantha*. In the protologue, Singer (1962) implied Singer B 2525 as holotype, not citing any other material. Later, Singer (1964) indicated that the collection under this number had a putative isotype at BAFC. However, the B 2525 holotype cannot be located at LIL (A. Hladki pers. corr.), and the B 2525 isotype collection cannot be located at BACF (S. Pereira, pers. corr.). On the other hand, Singer B 1997 and Singer B 2112 are the only specimens deposited in BAFC and LIL, respectively. They were collected in 1956 and identified by Singer himself, and, although no determination date is noted on the herbarium sheet label, we believe that they represent part of the original material of the species (McNeill et al. 2006: Art. 9.10).

The authors also asked whether any material labeled as *Oudemansiella macracantha* is available at F, FH, or MICH, institutions where Rolf Singer also worked and deposited materials (Mueller 1995, Mueller & Wu 1997). All responded that no materials exist under this name. The exsiccata of INPA are available at Species Link System < [www.splink.org.br/index](http://www.splink.org.br/index) > and no exsiccatum named *O. macracantha* is available in this herbarium. Thus, the authors choose here to designate Singer B 1997 (BAFC 51670) as the lectotype of *O. macracantha*.

*Oudemansiella glutinosa* Singer from Colombia, which also has ornamented basidiospores, differs from *O. macracantha* and *O. steffenii* in the gelatinized zones in pileus and stipe and the considerably smaller basidiospores (14–16.5 × 12–14 µm, including the 2–3 mm high ornamentation; Singer 1989). The basidiospores of *O. macracantha* and *O. steffenii* range more than 20 mm with ornamentations (Singer 1964).

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## MYCOTAXON

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***Disciseda bovista*, recently collected from northern Italy,  
and *Lycoperdon defossum*, a synonym of *D. candida***

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**Abstract** — The rare *Disciseda bovista* is described from northern Italy (Piedmont) based upon a recent collection. This is the second documented collection of the species from Italy, and the first in recent times. The study of Vittadini's original material labelled as *Lycoperdon defossum*, a taxon considered by many authors as a synonym of *D. bovista*, reveals that it should be ascribed to *D. candida* and, as such, represents the first record of the species from Italy.

**Key words** — *Agaricales*, *Lycoperdaceae*, *Catastoma*, red lists, taxonomy

**Introduction**

*Disciseda* Czern. (= *Catastoma* Morgan) is a genus belonging to the gasteroid lineage of the *Agaricaceae* Chevall. s.l. (Bates et al. 2009, Gube 2009), where it forms the *Disciseda*-clade (Larsson & Jeppson 2008), which is basal to the rest of the taxa formerly placed within the *Lycoperdaceae* Chevall. The genus has a worldwide distribution, but all the species are restricted to xeric habitats. In the latest edition of the DICTIONARY OF THE FUNGI (Kirk et al. 2008), fifteen *Disciseda* species are recognized.

The genus is characterized by semi-hypogeous basidiomes with a loose mycelial connection, and a peculiar type of dehiscence (e.g. Mattiolo 1934, Ahmad 1950, Mitchel et al. 1975, Jeppson 1997, Calonge 1998, Moreno et al. 2003). The ostiole develops in the basal zone of the endoperidium; then the exoperidium cracks along the circumference of the basidiome and the upper part gets detached from its hypogean portion. When disturbed by atmospheric agents, the detached basidiome will turn over and, consequently, exposes the basal portion of the endoperidium, which places the ostiole in the apical

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\* Corresponding author

position. The portion of the exoperidium that initially covered the apical parts of the semi-hypogeous basidiome remains attached to the base as a kind of cap encrusted with soil particles and vegetal debris. Kreisel (1962) termed these fungi ‘geanemochorous tumblers’, since the whole basidiome is blown by the wind, causing basidiospore dispersal.

This paper reports on the occurrence of the rare and threatened *Disciseda bovista* in Piedmont, northern Italy, and provides observations on previous Italian collections. Furthermore, we provide our analysis of Vittadini’s collection originally labelled *Lycoperdon defossum*, a taxon considered by some mycologists to represent a synonym of *D. bovista*.

## Material and methods

The macro- and micromorphological features are described from notes taken from fresh material. The micromorphological features were observed from dried material mounted in distilled water and Congo red. Spore size is expressed both as a range and mean value based on 26 randomly chosen basidiospores. Basidiospore measurements do not include either sterigma or ornamentation.

Author citations follow Index Fungorum (<http://www.indexfungorum.org/Names/AuthorsOfFungalNames.asp>). Herbarium abbreviations are according to Thiers (2010).

## Taxonomy

*Disciseda bovista* (Klotzsch) Henn., Beiblatt zur Hedwigia 42: 128, 1903,  
sub “Lloyd, G.G. *Catastoma*”.

FIGS. 1a–g

≡ *Geastrum bovista* Klotzsch, Nov. Actorum Acad. Caes. Leop.-  
Carol. Nat. Cur. 19(Suppl. 1): 243, 1843.

≡ *Catastoma bovista* (Klotzsch) Hollós, in Hennings, Verh.  
Bot. Vereins Prov. Brandenburg 43: VI, 1901.

= *Globaria debreceniensis* Hazsl., Verh. Zool.-Bot. Ges. Wien 26: 226, 1876.

≡ *Bovista debreceniensis* (Hazsl.) De Toni, Sylloge Fungorum 7: 476, 1888.

≡ *Catastoma debreceniense* (Hazsl.) Hollós, Termés. Közl. 56: 186, 1900.

≡ *Disciseda debreceniensis* (Hazsl.) Hollós, Termés. Fü. 25: 102, 1902.

= *Bovista subterranea* Peck, Bot. Gazette (Crawfordsville) 4(10): 216, 1879.

≡ *Catastoma subterraneum* (Peck) Morgan, J. Cincinnati Soc. Nat. Hist. 14: 143, 1892.

≡ *Disciseda subterranea* (Peck) Coker & Couch, Gast. East. U.S. and Canada: 141, 1928.

SELECTED DESCRIPTIONS: Kers (1975: 420–427); Calonge (1998: 79–80).

SELECTED ICONOGRAPHY: Mattiolo (1934: FIGS. 1–16); Kers (1975: FIG. 2); Jeppson (1997: FIG. 1); Jordal et al. (2007: FIG. 1).

BASIDIOME (8–)10–26 mm in diam. × 9–15 mm in height, globose, subglobose, regular to gibbous, sometimes lobed and depressed, mottled. Immature basidiomes completely enveloped by the exoperidium (Fig. 1a) resembling the protective cases of some trichopteran larvae. Mature basidiomes enveloped at the base by remnants of the exoperidium that forms a thick mycelial pad, heavily

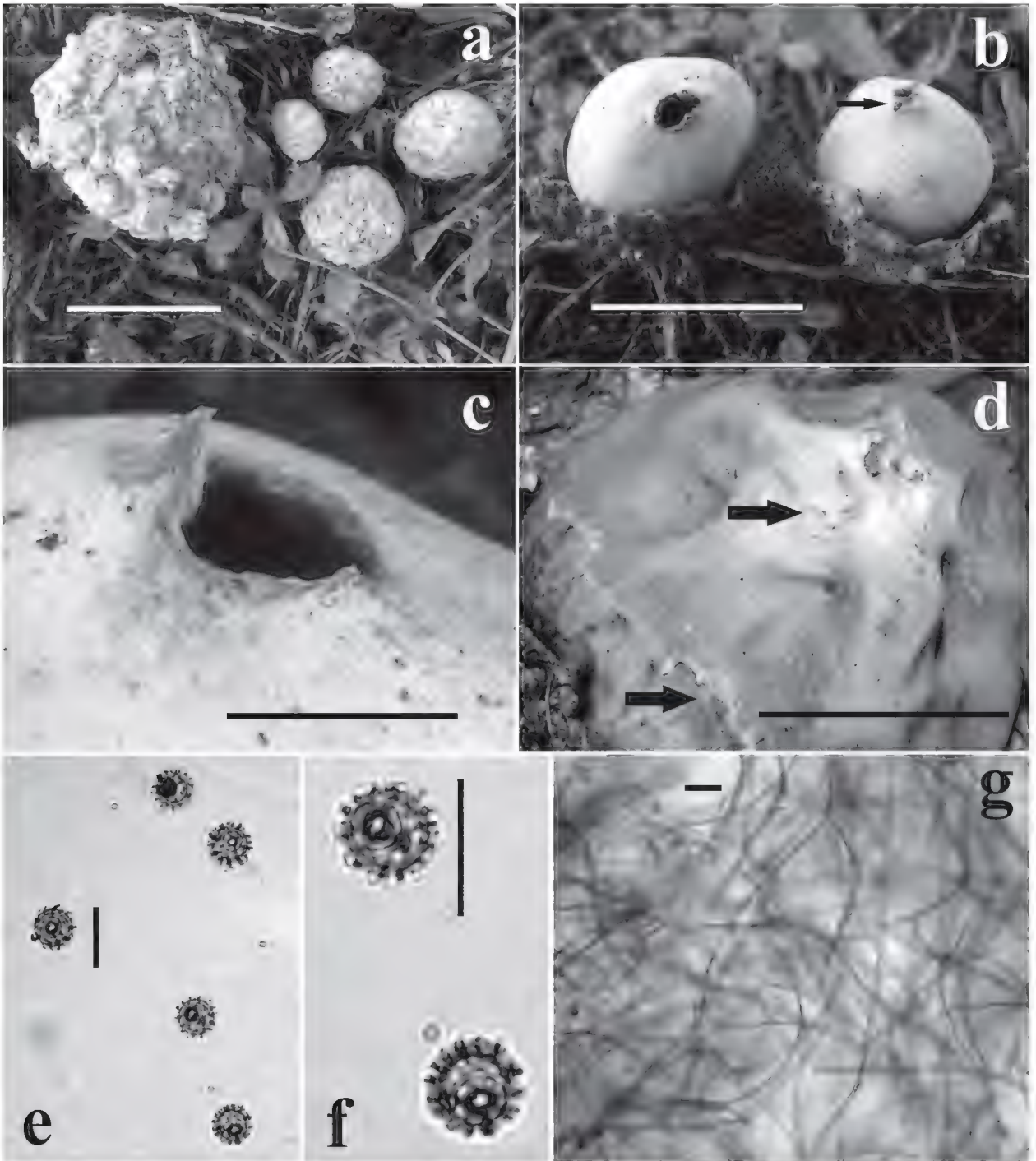


FIGURE 1. *Disciseda bovista* (TO HG1998). a. Immature basidiomes with intact and encrusted exoperidium. b. Ripe and inverted basidiomes with crumbling exoperidium; arrow = double ostiole. c. Ostiole. d. Mottled endoperidium lacking an evident ostiole, with remnants of the pseudoparenchymatic exoperidial layer (arrows). e, f. Basidiospores. g. Capillitium. Bars: a = 10 mm; b = 20 mm; c = 2mm; d = 20 mm; e, f = 10  $\mu$ m; g = 20  $\mu$ m.

encrusted with plant debris and particles of soil (FIG. 1b). Old basidiomes often colonized by green algae. OSTIOLE (1–)2 mm in diam., orbicular to irregularly-shaped and torn (FIG. 1c); in some ripe specimens no ostiole has been observed; occasionally additional little ostioles are present. EXOPERIDIUM colour very difficult to discern as the exoperidium is heavily encrusted with debris,



whitish to greyish brown. ENDOPERIDIUM white to light grey, then yellowish-brown, coriaceous, leathery, persistent, glabrous to rimulose-pubescent, often with small patches, remnants of the pseudoparenchymatic exoperidial layer (FIG. 1d). GLEBA light brown to dark brown, cottony at first, soon becoming pulverulent.

BASIDIOSPORES  $(5.8-6.0-7.4(-7.6) \times (5.5-5.8-7.0(-7.1) \mu\text{m}$ , on average  $6.68 \times 6.38 \mu\text{m}$ ,  $Q = 1.0-1.07$ ,  $Q_m = 1.046$  ( $n = 26$ ), globose, baculate, *Terfezia*-like, warts cylindrical or truncate-conical, up to  $0.5-1 \mu\text{m}$  long, yellowish brown in water mounts, with a central to eccentric large oil drop (FIG. 1e, f); sterigmal remnants (pedicels) short (up to  $2-3 \mu\text{m}$  in length) or absent. CAPILLITIUM of the *Lycoperdon*-type,  $2.5-5 \mu\text{m}$  in diam., with rounded tips, thick-walled (up to  $1.0 \mu\text{m}$ ), fragile, pale brown in water mounts, straight to undulate, rarely finely encrusted, sometimes with small-sized pores, septate, often disarticulating at the septum, occasionally with dichotomous branching (FIG. 1g). EXOPERIDIUM two-layered: (1) outer mycelial layer, with  $2-4 \mu\text{m}$  wide hyphal elements, interwoven with plant matter and particles of soil; (2) inner pseudoparenchymatous layer, gelatinous, up to  $1 \text{ mm}$  thick, made up of  $10-20 \mu\text{m}$  in diam., rounded, thin-walled cells. ENDOPERIDIUM consisting of  $2-5 \mu\text{m}$  wide, thin and thick walled hyphae.

HABITAT. Terrestrial, found in an ex-vineyard arid soil partially covered with xerophilic mosses.

MATERIAL EXAMINED – ITALY: Piedmont, Perosa Canavese (Torino),  $45^\circ 23' 55.19'' \text{ N}$ ,  $7^\circ 50' 02.14'' \text{ E}$ , 262 m a.s.l., 10 Dec. 2009, *legit* L. Panno, *det.* A. Vizzini (TO HG1998).

## Discussion

Distributed in Europe and America (Coker & Couch 1928, Calonge 1998, Kreisel 2001), where it typically grows in dry, sandy, sunlit and usually steppe-like habitats (Kers 1975, Calonge 1998, Jordal et al. 2007, Stasińska 2008), *D. bovista* is a rare gasteroid species that seems to be declining in Northern Europe. As a consequence, it has been included in the red-lists of rare and threatened macromycetes of several European countries (e.g. Switzerland, Senn-Irlet et al. 1997; The Netherlands, Arnolds & Kuyper 1996; Italy, Venturella et al. 1997; Austria, Krisai-Greilhuber 1999; Denmark, Stoltze & Pihl 1998; Poland, Wojewoda & Ławrynowicz 2004; Sweden, Gärdenfors 2005).

*Disciseda bovista* is characterized by the  $6-7 \mu\text{m}$  diam., strongly ornamented spores, with *Terfezia*-like, truncate-conical warts, and without long sterigmal remnants. Among the closest allies, *D. candida*, which macromorphologically may be easily confused with *D. bovista*, is clearly distinguished by the smaller ( $3.5-5.5 \mu\text{m}$ ), finely ornamented basidiospores (e.g. Kers 1975, Jeppson 1986, Mornand 1990, Moyersoen & Demoulin 1996, Calonge 1998, Poumarat et al.

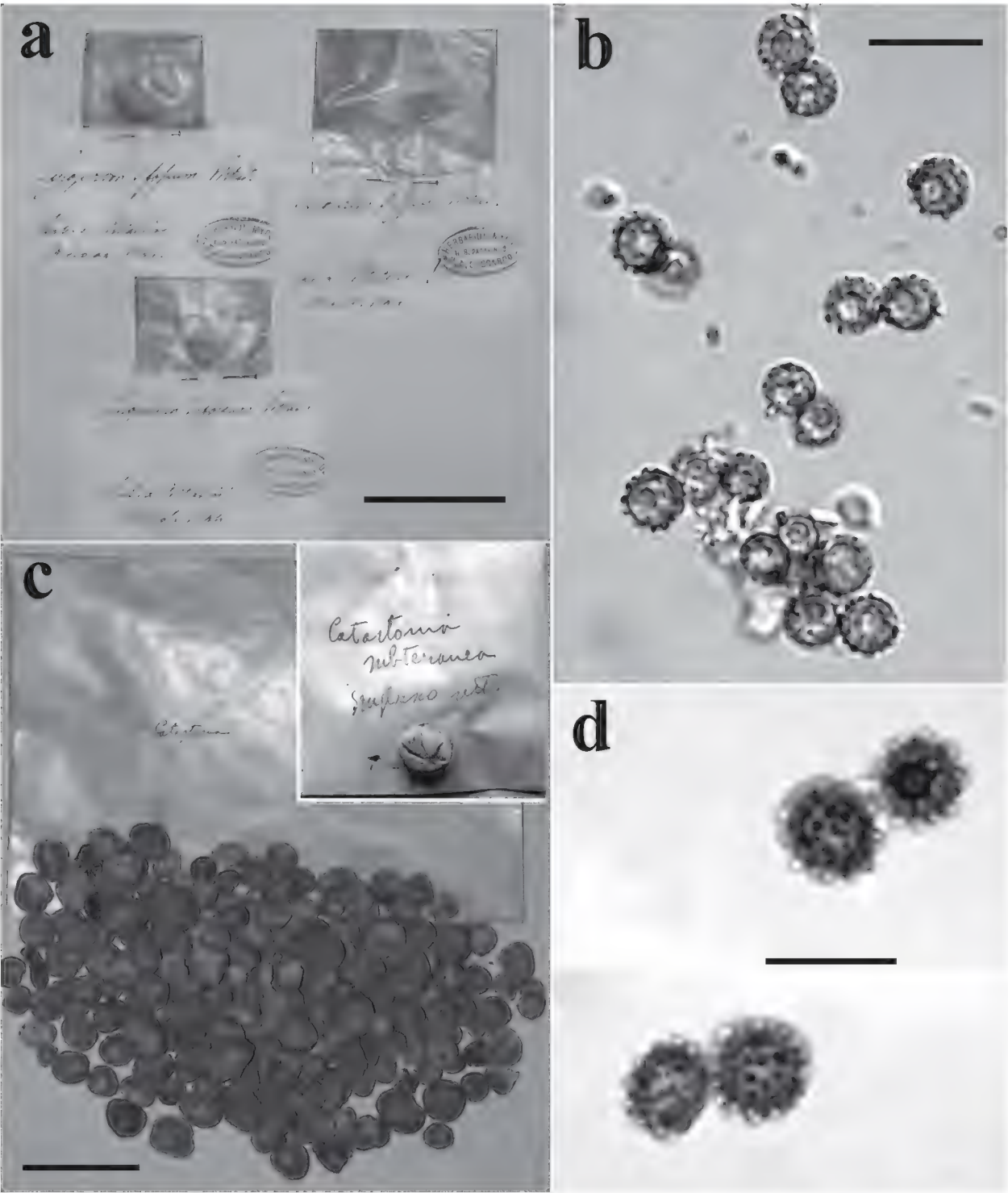


FIGURE 2. a–b. Vittadini’s collection (PAD) – *Lycoperdon defossum*. a. Basidiomes. b. Basidiospores. c–d. Mattirolò’s collection (TO) – *Catastoma subterraneum*. c. Basidiomes. d. Basidiospores. Bars: a, c = 5 cm; b, d = 10 µm.

2000, Sarasini 2005, Bates et al. 2009). *Disciseda cervina* (Berk.) Hollós has smaller (4.0–5.6 µm), smooth to asperulate basidiospores and an endoperidium often with purplish hues (Hollós 1903, Poumarat 2003, Bates et al. 2009), while the recently described *D. nigra* Dörfelt & H. Nowak from Germany differs in its blackish mature endoperidium and larger basidiospores (7.5–8.5 µm) with warts up to 1.8 µm high (Dörfelt & Nowak 2002).

As regards collections of *D. bovista* previously reported from Italy, the Italian Checklist (Onofri et al. 2005) mentions only one find from Sardinia (Brotzu 1994) included in a local checklist without any supporting data: no description, iconography, observations, or herbarium number were provided. Photos and description of the species, included in the monograph on epigeous gasteromycetes by the Italian specialist Sarasini (2005), refer to Spanish specimens.

Many authors consider *Lycoperdon defossum* Vittad. (Vittadini 1842) a synonym of *Disciseda bovista* [e.g. Petri 1909, Lloyd (Mattiolo 1934), Mattiolo 1934, Calonge 1998; but see Moravec 1958 and Sarasini 2005]. According to Stafleu & Cowan (1986), Vittadini's original specimens are preserved at TO and PAD, which we checked for *L. defossum* collections. The collection cited by Mattiolo (1934) and probably examined by Lloyd (Mattiolo 1934) is missing from TO, but we have located Vittadini's original material in Saccardo's herbarium (PAD). That collection consists of three pressed specimens from Milan (FIG. 2a) with 4–5.5 µm diam., finely to medium ornamented basidiospores (FIG. 2b) that clearly support the material in *D. candida*. This collection represents the first record of the species from Italy.

While studying and sorting out Mattiolo's herbarium of epi- and hypogeous gasteromycetoid fungi housed at TO, we were able to study a very large collection (consisting of over a hundred specimens, FIG. 2c) labelled as *Catastoma subterraneum* made in the Turin hinterland (Mattiolo 1934). Based on spore features, the collection also appears to represent *D. bovista* (FIG. 2d).

In conclusion, our paper describes the second collection — the first in recent times — of *Disciseda bovista* from Italy that can be documented with certainty. We also demonstrate that *Lycoperdon defossum* is not a synonym of *D. bovista*, but rather of *D. candida*:

***Disciseda candida*** (Schwein.) Lloyd, Mycol. Writ. 1: 100, 1902.

= *Lycoperdon defossum* Vittad., Monogr. Lycoperd.: 33, 1842, nom. illegit., non Batsch 1789.

= *Globaria defossa* Quél., Bull. Soc. Bot. France 24: 327, 1878, nom. nov.

= *Bovista defossa* (Quél.) De Toni, Syll. Fung. 7: 101, 1888.

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## HKU(M) moves to IFRDC Kunming

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**Abstract** — The University of Hong Kong Mycological Herbarium (HKU(M)) has been relocated to the International Fungal Research & Development Centre, Chinese Academy of Forestry, in Kunming, Yunnan, PR China. The official acronym of the herbarium is IFRD. New IFRD numbers for 30 generic types and 238 specific types transferred from HKU(M) are listed here.

**Key words** — tropical fungal specimens

## Introduction

Between 1993 and December 2006, K.D. Hyde and co-workers published 388 scientific papers describing 60 new genera, 411 new species, and numerous other collections of tropical fungi (e.g., Poon & Hyde 1998, Wong & Hyde 2001, Ho et al. 2002, Kumar & Hyde 2004). Most type material from these publications was deposited in the Herbarium of the University of Hong Kong (official acronym = HKU). However, the herbarium was unwilling to curate the fungal specimens due to lack of funding. Therefore, all fungal specimens were curated by Helen Y.M. Leung in a separate wing of the herbarium known as HKU(M). Than et al. (2008) and Tang et al. (2009) also deposited specimens in HKU(M).

K.D. Hyde, the mycologist at the University of Hong Kong, resigned from his post on 31 December 2007. The University did not replace him and had no interest in maintaining the fungal herbarium, which comprised 7934 mostly

tropical fungal specimens, including 60 generic types and 411 specific types. A decision was therefore made to move the herbarium to where it is now located in the International Fungal Research & Development Centre in Kunming, PR China. The new herbarium was registered with Index Herbariorum in early 2008 and has the official acronym IFRD. The curator is Dr. Hang Chen.

The purpose of this paper is to list the types of genera and species that have been moved to IFRD and accessioned with new numbers. TABLE 1 lists 241 new IFRD numbers with their HKU(M) equivalents.

Unfortunately, many specimens were misplaced during transit from Hong Kong to Kunming, and we are still searching for these specimens. In a later paper, we will list any additional types that have been located, including isotypes or paratypes (if available), for any whose holotypes are missing at the present time.

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TABLE 1. New IFRD numbers assigned to 30 generic types and 238 specific types.  
(All collections are holotypes, unless explicitly labelled as isotypes or paratypes.)

SPECIES	ACCESSION NUMBERS	
	ORIGINAL	NEW
<i>Acrodictys liputii</i> L. Cai et al.	HKU(M) 16490 [isotype]	IFRD 8640 [isotype]
<i>Acrogenospora ovalis</i> Goh et al.	HKU(M) 4743	IFRD 8641
<i>Acrogenospora verrucospora</i> Hong Zhu et al.	HKU(M) 17494	IFRD 8642
<i>Amarenographium sinense</i> Joanne E. Taylor et al.	HKU(M) 3989	IFRD 8643
<i>Aniptodera inflatiascigera</i> K.M. Tsui et al.	HKU(M) 4672	IFRD 8644
<i>Aniptodera intermedia</i> K.D. Hyde & Alias	HKU(M) 7468	IFRD 8645
<i>Aniptodera mauritaniensis</i> K.D. Hyde et al.	HKU(M) 2616	IFRD 8646
<i>Aniptodera palmicola</i> K.D. Hyde et al.	HKU(M) 2205	IFRD 8647
<i>Annulatascus joannae</i> K.M. Tsui et al.	HKU(M) 12177	IFRD 8648
<i>Annulatascus palmietensis</i> Goh et al.	HKU(M) 2206	IFRD 8649
<i>Annulatascus triseptatus</i> S.W. Wong et al.	HKU(M) 3129	IFRD 8650
<i>Anthostomella acuminata</i> B.S. Lu & K.D. Hyde	HKU(M) 2118	IFRD 8651
<i>Anthostomella applanata</i> B.S. Lu & K.D. Hyde	HKU(M) 2109	IFRD 8652
<i>Anthostomella aquatica</i> K.D. Hyde & Goh	HKU(M) 2774	IFRD 8653
<i>Anthostomella birima</i> Joanne E. Taylor et al.	HKU(M) 3909	IFRD 8654
<i>Anthostomella caffrariae</i> B.S. Lu & K.D. Hyde	HKU(M) 2140	IFRD 8655
<i>Anthostomella clypeosa</i> Joanne E. Taylor et al.	HKU(M) 3633	IFRD 8656
<i>Anthostomella colligata</i> K.D. Hyde & B.S. Lu	HKU(M) 2160	IFRD 8657
<i>Anthostomella frondicola</i> K.D. Hyde et al.	HKU(M) 535	IFRD 8658
<i>Anthostomella kapitiae</i> Whitton et al.	HKU(M) 5007	IFRD 8659
<i>Anthostomella longa</i> B.S. Lu et al.	HKU(M) 7311	IFRD 8660
<i>Anthostomella manawatua</i> Whitton et al.	HKU(M) 5008	IFRD 8661
<i>Anthostomella meerensis</i> B.S. Lu & K.D. Hyde	HKU(M) 2134	IFRD 8662
<i>Anthostomella notabilis</i> Joanne E. Taylor et al.	HKU(M) 4294	IFRD 8663
<i>Anthostomella nypae</i> K.D. Hyde et al.	HKU(M) 7305	IFRD 8664
<i>Anthostomella nypensis</i> K.D. Hyde et al.	HKU(M) 7306	IFRD 8665
<i>Anthostomella nypicola</i> K.D. Hyde et al.	HKU(M) 7307	IFRD 8666
<i>Anthostomella oblongata</i> B.S. Lu & K.D. Hyde	HKU(M) 7313	IFRD 8667
<i>Anthostomella okatina</i> Whitton et al.	HKU(M) 5013	IFRD 8668
<i>Anthostomella palmae</i> K.D. Hyde & B.S. Lu	HKU(M) 2209	IFRD 8669
<i>Anthostomella raphiae</i> B.S. Lu & K.D. Hyde	HKU(M) 2146	IFRD 8670
<i>Anthostomella spiralis</i> K.D. Hyde & B.S. Lu	HKU(M) 2119	IFRD 8671
<i>Anthostomella variabilis</i> B.S. Lu & K.D. Hyde	HKU(M) 2091	IFRD 8672
<i>Anthostomella xuanenensis</i> Joanne E. Taylor et al.	HKU(M) 4064	IFRD 8673
<i>Apioclypea cocoicola</i> Joanne E. Taylor et al.	HKU(M) 4344	IFRD 8674
<i>Apioclypea nonapiospora</i> Joanne E. Taylor et al.	HKU(M) 3657	IFRD 8675



<i>Apioclypea nypicola</i> K.D. Hyde et al.	HKU(M) 1629	IFRD 8676
<i>Apioclypea phoenicicola</i> K.D. Hyde et al.	HKU(M) 1665	IFRD 8677
<i>Apiospora sinensis</i> K.D. Hyde et al.	HKU(M) 3963	IFRD 8678
<i>Appendicospora hongkongensis</i> Yanna et al.	HKU(M) 5301	IFRD 8679
<i>Aqualignicola hyalina</i> Ranghoo et al. [generic type]	HKU(M) 12178	IFRD 8680
<i>Aquaphila albicans</i> Goh et al. [generic type]	HKU(M) 2856	IFRD 8681
<i>Aquaticola longicolla</i> K.M. Tsui et al.	HKU(M) 5159	IFRD 8682
<i>Aquaticola minutiguttulata</i> K.M. Tsui et al.	HKU(M) 12275	IFRD 8683
<i>Aquaticola triseptata</i> K.M. Tsui et al.	HKU(M) 12222	IFRD 8684
<i>Arecacicola calami</i> Joanne E. Taylor et al. [generic type]	HKU(M) 1126a	IFRD 8685
<i>Arecomyces bruneiensis</i> K.D. Hyde	HKU(M) 1728	IFRD 8686
<i>Arecomyces dicksonii</i> K.D. Hyde	HKU(M) 2641	IFRD 8687
<i>Arecomyces epigenus</i> K.D. Hyde	HKU(M) 1529	IFRD 8688
<i>Arecomyces frondicola</i> K.D. Hyde [generic type]	HKU(M) 1713	IFRD 8689
<i>Arecomyces hedgeri</i> K.D. Hyde	HKU(M) 2685	IFRD 8690
<i>Arecomyces sekoyae</i> K.D. Hyde	HKU(M) 2682	IFRD 8691
<i>Arecomyces tetrasporus</i> K.D. Hyde	HKU(M) 2714	IFRD 8692
<i>Ascolacicola aquatica</i> Ranghoo & K.D. Hyde [generic type]	HKU(M) 5243	IFRD 8693
<i>Ascomauritiana lignicola</i> Ranghoo & K.D. Hyde [generic type]	HKU(M) 5255	IFRD 8694
<i>Ascominuta lignicola</i> Ranghoo & K.D. Hyde [generic type]	HKU(M) 5246	IFRD 8695
<i>Ascotaiwania mitriformis</i> Ranghoo & K.D. Hyde	HKU(M) 5224	IFRD 8696
<i>Ascotaiwania pallida</i> K.D. Hyde & Goh	HKU(M) 3247	IFRD 8697
<i>Astrocystis nypae</i> G.J.D. Sm. & K.D. Hyde	HKU(M) 1626	IFRD 8698
<i>Astrocystis selangorensis</i> G.J.D. Sm. & K.D. Hyde	HKU(M) 1625a	IFRD 8699
<i>Astrocystis sinensis</i> Joanne E. Taylor et al.	HKU(M) 4087	IFRD 8700
<i>Astrosphaeriella aequatoriensis</i> K.D. Hyde & J. Fröhl.	HKU(M) 2707	IFRD 8701
<i>Astrosphaeriella immersa</i> Joanne E. Taylor et al.	HKU(M) 3683	IFRD 8702
<i>Astrosphaeriella lenticularis</i> K.D. Hyde & J. Fröhl.	HKU(M) 2733	IFRD 8703
<i>Astrosphaeriella papillata</i> K.D. Hyde & J. Fröhl	HKU(M) 2018	IFRD 8704
<i>Astrosphaeriella splendida</i> K.D. Hyde & J. Fröhl.	HKU(M) 2732	IFRD 8705
<i>Ayria appendiculata</i> Fryar & K.D. Hyde [generic type]	HKU(M) 15553	IFRD 8706
<i>Balaniopsis dendroidea</i> Whitton et al.	HKU(M) 5098	IFRD 8707
<i>Balaniopsis kirkii</i> Whitton et al.	HKU(M) 5099	IFRD 8708
<i>Boerlagiomyces grandisporus</i> S.J. Stanley & K.D. Hyde	HKU(M) 2978	IFRD 8709
<i>Botryosphaeria archontophoenicis</i> Joanne E. Taylor et al.	HKU(M) 3539	IFRD 8710
<i>Botryosphaeria brunneispora</i> Joanne E. Taylor et al.	HKU(M) 3987	IFRD 8711
<i>Brachydesmiella verrucosa</i> Goh et al.	HKU(M) 5855	IFRD 8712

<i>Brachysporiopsis chinensis</i> Yanna et al. [generic type]	HKU(M) 13660	IFRD 8713
<i>Brunneiapiospora aequatoriensis</i> K.D. Hyde et al.	HKU(M) 2656	IFRD 8714
<i>Brunneiapiospora daemonoropis</i> K.D. Hyde et al.	HKU(M) 1974	IFRD 8715
<i>Brunneiapiospora javensis</i> K.D. Hyde et al. [generic type]	HKU(M) 1121b	IFRD 8716
<i>Brunneospora aquatica</i> Ranghoo & K.D. Hyde [generic type]	HKU(M) 5251	IFRD 8717
<i>Camposporium fusisporum</i> Whitton et al.	HKU(M) 12925	IFRD 8718
<i>Camposporium ramosum</i> Whitton et al.	HKU(M) 12924	IFRD 8719
<i>Canalisporium exiguum</i> Goh & K.D. Hyde	HKU(M) 3349	IFRD 8720
<i>Canalisporium variabile</i> Goh & K.D. Hyde	HKU(M) 7438	IFRD 8721
<i>Cancellidium pinicola</i> S.Y. Yeung et al.	HKU(M) 17167	IFRD 8722
<i>Capsulospora trachycarpa</i> Joanne E. Taylor et al.	HKU(M) 3986	IFRD 8723
<i>Cataractispora appendiculata</i> K.D. Hyde et al.	HKU(M) 3120	IFRD 8724
<i>Cataractispora aquatica</i> K.D. Hyde et al. [generic type]	HKU(M) 3123	IFRD 8725
<i>Cataractispora viscosa</i> K.D. Hyde et al.	HKU(M) 3130	IFRD 8726
<i>Ceuthospora palmicola</i> Joanne E. Taylor et al.	HKU(M) 4125	IFRD 8727
<i>Chaetopsina alexandrae</i> Joanne E. Taylor et al.	HKU(M) 3623	IFRD 8728
<i>Chaetopsina hongkongensis</i> Goh & K.D. Hyde	HKU(M) 2623	IFRD 8729
<i>Chloridium cocoicola</i> Joanne E. Taylor et al.	HKU(M) 4228	IFRD 8730
<i>Circinotrichum palmicola</i> Joanne E. Taylor et al.	HKU(M) 3933	IFRD 8731
<i>Clohiesia lignicola</i> K.M. Tsui et al.	HKU(M) 5539	IFRD 8732
<i>Clypeosphaeria uniseptata</i> K.M. Tsui et al.	HKU(M) 8095	IFRD 8733
<i>Cordana uniseptata</i> L. Cai et al.	HKU(M) 17163	IFRD 8734
<i>Curvatispora singaporensis</i> V.V. Sarma & K.D. Hyde [generic type]	HKU(M) 12457	IFRD 8735
<i>Cytoplacosphaeria phragmiticola</i> Poon & K.D. Hyde	HKU(M) 5191	IFRD 8736
<i>Dactylaria biguttulata</i> Goh & K.D. Hyde	HKU(M) 3334	IFRD 8737
<i>Dactylaria hyalotunicata</i> K.M. Tsui et al.	HKU(M) 5377	IFRD 8738
<i>Dactylaria lakebarrinensis</i> Goh & K.D. Hyde	HKU(M) 3162	IFRD 8739
<i>Dactylaria obscuriseptata</i> Goh & K.D. Hyde	HKU(M) 4738(PC7)	IFRD 8740a
<i>Dactylaria plovercovensis</i> Goh & K.D. Hyde	HKU(M) 4738(PC7A)	IFRD 8740b
<i>Diaporthe palmarum</i> Joanne E. Taylor et al.	HKU(M) 4064	IFRD 8741
<i>Dictyochaeta microcylindrospora</i> Whitton et al.	HKU(M) 4932	IFRD 8742
<i>Dictyochaeta multisetula</i> Whitton et al.	HKU(M) 4922	IFRD 8743
<i>Dictyochaeta plovercovensis</i> Goh & K.D. Hyde	HKU(M) 4724	IFRD 8744
<i>Dictyochaeta renispora</i> Whitton et al.	HKU(M) 4914	IFRD 8745
<i>Dictyochaeta seychellensis</i> Whitton et al.	HKU(M) 4926	IFRD 8746
<i>Dictyosporium canisporum</i> L. Cai & K.D. Hyde	HKU(M) 17156	IFRD 8747
<i>Dictyosporium giganticum</i> Goh & K.D. Hyde	HKU(M) 2184	IFRD 8748
<i>Dictyosporium tetraploides</i> L. Cai & K.D. Hyde	HKU(M) 17146	IFRD 8749

<i>Dictyosporium tetraseriale</i> Goh et al.	HKU(M) 5327	IFRD 8750
<i>Digitodesmium heptasporum</i> L. Cai & K.D. Hyde	HKU(M) 17158	IFRD 8751
<i>Elegantimycetes sporidesmiopsis</i> Goh et al. [generic type]	HKU(M) 5378	IFRD 8752
<i>Endomelanconium phoenicicola</i> Yanna et al.	HKU(M) 10023	IFRD 8753
<i>Endophragmiella bitriseptata</i> Goh et al.	HKU(M) 5184	IFRD 8754
<i>Endophragmiella triseptata</i> K.M. Tsui et al.	HKU(M) 5402	IFRD 8755
<i>Endosporoideus pedicellatus</i> W.H. Ho et al. [generic type]	HKU(M) 10066	IFRD 8756
<i>Everhartia phoenicis</i> Yanna et al.	HKU(M) 10504	IFRD 8757
<i>Fluviatispora boothii</i> Fryar & K.D. Hyde	HKU(M) 15792	IFRD 8758
<i>Frigidispora colnensis</i> K.D. Hyde & Goh [generic type]	HKU(M) 3244	IFRD 8759
<i>Frondisphaeria joanneae</i> J. Fröhl. & K.D. Hyde	HKU(M) JF 718	IFRD 8760
<i>Guestia gonetropospora</i> G.J.D. Sm. & K.D. Hyde [generic type]	HKU(M) 3347	IFRD 8761
<i>Halosarpheia heteroguttulata</i> S.W. Wong et al.	HKU(M) 2806	IFRD 8762
<i>Helicorhoidion nypicola</i> K.D. Hyde & Goh	HKU(M) 8276	IFRD 8763
<i>Helicosporium gigasporum</i> K.M. Tsui et al.	HKU(M) 8091	IFRD 8764
<i>Herpotrichia dalisayi</i> K.D. Hyde & Aptroot	HKU(M) 2582	IFRD 8765
<i>Herpotrichia nypicola</i> K.D. Hyde & Alias	HKU(M) 5192	IFRD 8766
<i>Hydropisphaera ciliata</i> Joanne E. Taylor et al.	HKU(M) 3581	IFRD 8767
<i>Jahnula granulosa</i> K.D. Hyde & S.W. Wong	HKU(M) 2167	IFRD 8768
<i>Jahnula poonythii</i> K.D. Hyde & S.W. Wong	HKU(M) 2396	IFRD 8769
<i>Jahnula seychellensis</i> K.D. Hyde & S.W. Wong	HKU(M) 3239	IFRD 8770
<i>Jobellisia viridifusca</i> K.M. Tsui & K.D. Hyde	HKU(M) 8045	IFRD 8771
<i>Kionochaeta australiensis</i> Goh & K.D. Hyde	HKU(M) 2308	IFRD 8772
<i>Koorchaloma novojournalis</i> Yanna et al.	HKU(M) 7436	IFRD 8773
<i>Koorchaloma spartinicola</i> V.V. Sarma et al.	HKU(M) 10378	IFRD 8774
<i>Lachnum cylindricum</i> W.Y. Zhuang & K.D. Hyde	HKU(M) 10356	IFRD 8775
<i>Lachnum granulatum</i> W.Y. Zhuang et al.	HKU(M) 7177	IFRD 8776
<i>Lanceispora phyllophila</i> V.V. Sarma & K.D. Hyde	HKU(M) 8298	IFRD 8777
<i>Lasiosphaeria alexandrae</i> Joanne E. Taylor et al.	HKU(M) 3521	IFRD 8778
<i>Lasiosphaeria alexandricola</i> Joanne E. Taylor et al.	HKU(M) 3667	IFRD 8779
<i>Lasiosphaeria chapmanii</i> Joanne E. Taylor et al.	HKU(M) 7867	IFRD 8780
<i>Leptosphaeria ginimia</i> K.M. Tsui et al.	HKU(M) 16115	IFRD 8781
<i>Leptosphaeria nypicola</i> K.D. Hyde & Alias	HKU(M) 7160	IFRD 8782
<i>Lignincola nypae</i> K.D. Hyde & Alias	HKU(M) 6521	IFRD 8783
<i>Linocarpon alpiniae</i> K.D. Hyde	HKU(M) 1632	IFRD 8784
<i>Linocarpon apiculatum</i> K.D. Hyde	HKU(M) 1213	IFRD 8785
<i>Linocarpon appendisporum</i> K.D. Hyde	HKU(M) 1209	IFRD 8786
<i>Linocarpon aquaticum</i> K.D. Hyde	HKU(M) 1545	IFRD 8787
<i>Linocarpon australiense</i> K.D. Hyde	HKU(M) 1056	IFRD 8788

<i>Linocarpon breve</i> K.D. Hyde	HKU(M) 1097	IFRD 8789
<i>Linocarpon carinisorum</i> K.D. Hyde	HKU(M) 1830	IFRD 8790
<i>Linocarpon falciformisorum</i> K.D. Hyde	HKU(M) 1212	IFRD 8791
<i>Linocarpon lammiae</i> Whitton et al.	HKU(M) 12707	IFRD 8792
<i>Linocarpon luteocollum</i> Joanne E. Taylor et al.	HKU(M) 3573	IFRD 8793
<i>Linocarpon zingiberacicola</i> K.D. Hyde	HKU(M) 1920	IFRD 8794
<i>Lophiostoma maquilungense</i> K.D. Hyde & Aptroot	HKU(M) 2580	IFRD 8795
<i>Mangrovispora irregularis</i> Yanna et al.	HKU(M) 10877	IFRD 8796
<i>Massarina beaurivagea</i> Poonyth et al.	HKU(M) 10262	IFRD 8797
<i>Massarina grandispora</i> Joanne E. Taylor et al.	HKU(M) 4097	IFRD 8798
<i>Massarina mauritiana</i> Poonyth et al.	HKU(M) 10239	IFRD 8799
<i>Massarina phragmiticola</i> Poon & K.D. Hyde	HKU(M) 5188	IFRD 8800
<i>Massarina proprietunicata</i> K.M. Tsui et al.	HKU(M) 8058	IFRD 8801
<i>Massarina rhizophorae</i> Poonyth et al.	HKU(M) 10208	IFRD 8802
<i>Massarina sanguineo-ostiolata</i> Aptroot et al.	HKU(M) 7879	IFRD 8803
<i>Massarina thalassioidea</i> K.D. Hyde & Aptroot	HKU(M) 1882 [paratype]	IFRD 8804 [paratype]
<i>Mauritiana rhizophorae</i> Poonyth et al. [generic type]	HKU(M) 10219	IFRD 8805
<i>Menisporopsis multisetulata</i> K.M. Tsui et al.	HKU(M) 4662	IFRD 8806
<i>Monochaetiopsis lakefuxianensis</i> L. Cai et al. [generic type]	HKU(M) 8280	IFRD 8807
<i>Monodictys melanocephaloides</i> Goh & K.D. Hyde	HKU(M) 3334	IFRD 8808
<i>Monodictys trichocladopsis</i> Goh & K.D. Hyde	HKU(M) 4739	IFRD 8809
<i>Monotosporella sphaerica</i> Yanna & K.D. Hyde	HKU(M) 13287	IFRD 8810
<i>Muyocopron hongkongense</i> Joanne E. Taylor et al.	HKU(M) 3684	IFRD 8811
<i>Nemania saladerana</i> G.J.D. Sm. & K.D. Hyde	HKU(M) 3348	IFRD 8812
<i>Neolinocarpon enshiense</i> K.D. Hyde et al.	HKU(M) 3989	IFRD 8813
<i>Neolinocarpon inconspicuum</i> K.D. Hyde et al.	HKU(M) 3564	IFRD 8814
<i>Neolinocarpon nonappendiculatum</i> K.D. Hyde et al.	HKU(M) 3505	IFRD 8815
<i>Neolinocarpon nypicola</i> K.D. Hyde & Alias	HKU(M) 6518	IFRD 8816
<i>Niesslia palmicola</i> K.D. Hyde et al.	HKU(M) 4257	IFRD 8817
<i>Nigromammilla calami</i> K.D. Hyde & J. Fröhl. [generic type]	HKU(M) JF 863	IFRD 8818
<i>Ophioceras guttulatum</i> K.M. Tsui et al.	HKU(M) 12171	IFRD 8819
<i>Ophioceras hongkongense</i> K.M. Tsui et al.	HKU(M) 12226	IFRD 8820
<i>Ornatipora palmicola</i> K.D. Hyde et al. [generic type]	HKU(M) 2684	IFRD 8821
<i>Oxydothis bambusicola</i> Shenoy et al.	HKU(M) 17480	IFRD 8822
<i>Oxydothis ianei</i> Joanne E. Taylor et al.	HKU(M) 4060	IFRD 8823
<i>Palmicola bipolaris</i> Joanne E. Taylor et al.	HKU(M) 3823	IFRD 8824
<i>Paraceratocladium malaysianum</i> Goh & K.D. Hyde	HKU(M) 5854	IFRD 8825
<i>Parahendersonia trachycarpa</i> Joanne E. Taylor et al.	HKU(M) 4141	IFRD 8826



<i>Paraniesslia tuberculata</i> K.M. Tsui et al. [generic type]	HKU(M) 4647	IFRD 8827
<i>Periconia trachycarpicola</i> Joanne.E. Taylor et al.	HKU(M) 3957	IFRD 8828
<i>Phaeophleospora striae</i> Joanne E. Taylor et al.	HKU(M) 4127	IFRD 8829
<i>Phialogeniculata africana</i> Goh et al.	HKU(M) 2122	IFRD 8830
<i>Phomatospora archontophoenicis</i> Joanne E. Taylor et al.	HKU(M) 3641	IFRD 8831
<i>Phomatospora nypicola</i> K.D. Hyde & Alias	HKU(M) 7468	IFRD 8832
<i>Phragmitensis ellipsoidea</i> M.K.M. Wong et al.	HKU(M) 8001	IFRD 8833
<i>Podosporium biseptatum</i> Joanne E. Taylor et al.	HKU(M) 4235	IFRD 8834
<i>Polybulbophiale palmicola</i> Goh & K.D. Hyde [generic type]	HKU(M) 4717	IFRD 8835
<i>Polytretophora macrospora</i> Whitton et al.	HKU(M) 14024	IFRD 8836
<i>Porosphaerellopsis bipolaris</i> K.M. Tsui & K.D. Hyde	HKU(M) 12397	IFRD 8837
<i>Pseudohalonectria fuxianii</i> L. Cai et al.	HKU(M) 16126	IFRD 8838
<i>Pseudohalonectria miscanthicola</i> Shenoy et al.	HKU(M) 17487	IFRD 8839
<i>Pyricularia oncosperma</i> Yanna et al.	HKU(M) 10174	IFRD 8840
<i>Quintaria aquatica</i> K.D. Hyde & Goh	HKU(M) 848	IFRD 8841
<i>Ramichloridium lignicola</i> K.M. Tsui et al.	HKU(M) 12271	IFRD 8842
<i>Rivulicola aquatica</i> Ranghoo & K.D. Hyde	HKU(M) 5214	IFRD 8843
<i>Roussoella angustispora</i> D.Q. Zhou et al.	HKU(M) 9144	IFRD 8844
<i>Roussoella saltuensis</i> K.D. Hyde	HKU(M) 2717	IFRD 8845
<i>Saccardoella aquatica</i> K.M. Tsui et al.	HKU(M) 5371	IFRD 8846
<i>Saccardoella minuta</i> L. Cai & K.D. Hyde [isotype]	HKU(M) 17102	IFRD 8847 [isotype]
<i>Septoriella trachycarpi</i> Joanne E. Taylor et al.	HKU(M) 3986	IFRD 8848
<i>Sorokina frondicola</i> Joanne E. Taylor et al.	HKU(M) 3626	IFRD 8849
<i>Spadicoides minuta</i> L. Cai et al.	HKU(M) 17165	IFRD 8850
<i>Spadicoides palmicola</i> Goh & K.D.Hyde	HKU(M) 4785	IFRD 8851
<i>Spirodecospora bambusicola</i> B.S. Lu et al. [generic type]	HKU(M) 7303	IFRD 8852
<i>Sporoschisma paricuneatum</i> Goh & K.D. Hyde	HKU(M) 2550	IFRD 8853
<i>Stachybotrys reniverrucosa</i> Whitton et al.	HKU(M) 13093	IFRD 8854
<i>Stachybotrys waitakere</i> Whitton et al.	HKU(M) 13099	IFRD 8855
<i>Staurophoma calami</i> Yanna et al.	HKU(M) 7156	IFRD 8856
<i>Stictis ecclesiensis</i> Joanne E. Taylor et al.	HKU(M) 4042	IFRD 8857
<i>Stratiphoromyces brunneisorus</i> Goh & K.D. Hyde [generic type]	HKU(M) 4779	IFRD 8858
<i>Striatodecospora bambusae</i> D.Q. Zhou et al. [generic type]	HKU(M) 9143	IFRD 8859
<i>Submersisphaeria bambusicola</i> D.Q. Zhou & K.D. Hyde	HKU(M) 9045	IFRD 8860
<i>Sungaiicola bactrodesmiella</i> Fryar & K.D. Hyde [generic type]	HKU(M) 15201	IFRD 8861
<i>Tamsiniella labiosa</i> S.W. Wong et al. [generic type]	HKU(M) 2276	IFRD 8862
<i>Torrentispora crassiparietis</i> Fryar & K.D. Hyde	HKU(M) 15667	IFRD 8863

<i>Torrentispora fusiformis</i> Fryar & K.D. Hyde	HKU(M) 16048	IFRD 8864
<i>Triadelphia australiensis</i> Joanne E. Taylor et al.	HKU(M) 3587	IFRD 8865
<i>Tribulatia appendicospora</i> Joanne E. Taylor et al. [generic type]	HKU(M) 3620	IFRD 8866
<i>Trichocladium englandense</i> K.D. Hyde & Goh	HKU(M) 3255	IFRD 8867
<i>Trichocladium nypae</i> K.D. Hyde & Goh	HKU(M) 8276	IFRD 8868
<i>Vibrissea nypicola</i> K.D. Hyde & Alias	HKU(M) 6519	IFRD 8869
<i>Vismaya chaturbeeja</i> V.V. Sarma & K.D. Hyde [generic type]	HKU(M) 12457	IFRD 8870
<i>Wardomycopsis trachycarpicola</i> Joanne E. Taylor et al.	HKU(M) 16496	IFRD 8871
<i>Xylaria queenslandica</i> Joanne E. Taylor et al.	HKU(M) 3564	IFRD 8872
<i>Xylomyces giganteus</i> Goh et al.	HKU(M) 3188	IFRD 8873
<i>Xylomyces pusillus</i> Goh et al.	HKU(M) 4614	IFRD 8874
<i>Zygosporium pacificum</i> Whitton et al.	HKU(M) 12914	IFRD 8875
<i>Zygosporium pandanicola</i> Whitton et al.	HKU(M) 12919	IFRD 8876

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## MYCOTAXON

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**Studies on *Microthyriaceae*: some excluded genera**HAIXIA WU<sup>1\*</sup>, YANMEI LI<sup>1</sup>, HANG CHEN<sup>1</sup> & KEVIN D. HYDE<sup>1, 2, 3</sup><sup>\*</sup>Haixiawu1983@126.com<sup>1</sup> International Fungal Research and Development Centre,  
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**Abstract** — The genera *Asteronia*, *Dictyoasterina*, *Helminthopeltis*, and *Hidakaea* are presently included in the *Microthyriaceae*. We have examined the types, and their characters do not agree with this familial placement. We redescribe these four poorly known genera and suggest a more appropriate placement of these genera. *Asteronia* produces subglobose ascomata that develop on dark mycelium with copious hyphopodia on the host surface and should be placed in the *Asterinaceae* or *Meliolaceae*. *Dictyoasterina* has a black mycelial net, superficial hyphae with lateral hyphopodia, and globose ascomata with an ostiole and can be placed in *Asterinaceae*. The monotypic *Helminthopeltis*, characterized by elongated longitudinally cleft ascomata and filiform hyaline one-celled ascospores, should be placed in *Rhytismataceae*; *H. almeidaeana* is probably a synonym of a species in *Lophodermium*. *Hidakaea* has brown ascomata and unitunicate asci and should be considered in *Hypocreales*.

**Key words** — *Asteronia sweetiae*, *Dictyoasterina conopharyngiae*, *Helminthopeltis almeidaeana*, *Hidakaea tumidula*, taxonomy

**Introduction**

We are conducting studies on the *Dothideomycetes* in order to provide a natural classification (Zhang et al. 2008, 2009). As part of this work we are restudying the type species of the 49 genera placed in the *Microthyriaceae*, a poorly known but interesting family within the *Dothideomycetes* (Lumbsch & Huhndorf 2007). The important morphological characters of *Microthyriaceae* are superficial, flattened, dimidiate ascomata, the cells of upper wall of which are organized in



a radiating pattern, while a lower peridium is generally lacking. Members have a central ostiolar opening, cylindrical or broadly clavate to saccate, bitunicate and fissitunicate asci and ascospores that are hyaline or brown (Ryan 1924, 1926; Kirk et al. 2008). We have thus far examined several taxa within *Microthyriaceae* and in this paper report on four poorly known genera and their types: *Asteronia* (Hennings 1895) represented by *Asteronia sweetiae*, *Dictyoasterina* (Hansford 1947) represented by *Dictyoasterina conopharyngiae*, *Helminthopeltis* (Sousa da Câmara 1950) represented by *Helminthopeltis almeidaeana*, and *Hidakaea* (Hino & Katumoto 1955) represented by *Hidakaea tumidula*. Full descriptions of these taxa and suggestion for their taxonomic placement are provided.

### Materials and methods

Type specimens of *Asteronia sweetiae*, *Dictyoasterina conopharyngiae*, *Helminthopeltis almeidaeana*, and *Hidakaea tumidula* were obtained from K, IMI, LISE and YAM, respectively. Ascomata were rehydrated in 3% KOH prior to examination and sectioning. Specimens were examined under a stereo microscope (Leica MZ16A) and fine forceps were used to remove one or two ascomata, which were mounted in water, Melzer's, Congo red, or cotton blue reagents. Observations and photographs were made under the light microscopes (Nikon E800 and Leica DM3000). For some hyaline structures differential interference contrast microscopy was used.

Hand sections were cut with a sharp razor blade and thin (8 µm) sections were cut using a Leica CM1100 freezing microtome. The sections were transferred to a drop of water or a drop of cotton blue for examination and photography.

### Taxonomy

***Asteronia*** (Sacc.) Henn., Hedwigia 34: 104, 1895.

FIG. 1A–I

TYPE SPECIES: *Asteronia sweetiae* Henn., Hedwigia 34: 104, 1895.

≡ *Parodiopsis sweetiae* (Henn.) G. Arnaud, Annales des Épiphyties. 7: 53, 1921.

Colonies forming darkened regions on the underside of leaves, resembling a “sooty mold” to the unaided eye. Ascomata 130–180 µm high × 110–140 µm diam, gregarious, or some scattered, superficial, subglobose to globose, subcoriaceous, brown to black–brown, with an indistinct ostiole (FIGS 1A, B, C). Peridium 17–24 µm wide, one layered, composed of black-brown isodiametric cells of *textura angularis* (FIGS 1D, E). Hamathecium not apparent. Asci 60–93 × 20–27 µm (mean = 72 × 22.8 µm), 8-spored, bitunicate, oblong to broadly cylindrical, with a short pedicel, 8–9 µm long, 6.5–7.5 µm wide, apically rounded with an ocular chamber (FIGS 1F, H). Ascospores 33–46 × 4–6.5 µm (mean = 40.8 × 5.4 µm), tri-seriate to multiseriate, guttulate, thin-walled, straight or slightly curved, fusoid-ellipsoidal, widest in the middle part of the apical cell, with broadly rounded apex and tapering to a narrowly rounded base, hyaline, 1-septate, septum nearly central but nearer to apex, wall rough (FIGS 1G, I).



FIG. 1. *Asteronia sweetiae* (from holotype). A. Colonies of fungus on underside of host leaves. B. Appearance of ascomata on the host surface. C. Squash mount of ascoma. D, E. Section of ascomata. Note peridium. F, H. Asci Note the pedicel and ocular chamber. G, I. Ascospores. Scale bars: B = 200µm, C = 20 µm, D–I = 10 µm.

SPECIMEN EXAMINED: BRAZIL, Estado de Minas Gerais; on leaves of *Sweetia* sp. (*Fabaceae*), June 1892, E. Ulé (1968) (K (M) 159800, holotype).

*Asteronia* was described as a monotypic genus represented by *Asteronia sweetiae*. Subsequently, Hennings (1908) added *A. lauraceae* Henn. to the

genus. Two other names listed under *Asteronia* by Index Fungorum are errors for *Asterina appendiculosa* (Mont. & Berk.) Mont. and *Asterina erysiphoides* Kalchbr. & Cooke, species that were not included in *Asteronia* by Hennings (1895, 1908) and which have not been accepted or recombined by subsequent authors. Saccardo & Sydow (1899: 693) described *A. sweetiae* as parasitic with mycelium on the lower surface of leaves, ascomata that are subglobose and gregarious, and asci that are bitunicate with 8 spores. *Asteronia* is currently placed in the family *Microthyriaceae* (Lumbsch & Huhndorf 2007). However, the familial classification of *Asteronia* has been long confused. Hennings (1895) placed *Asteronia* in *Perisporiaceae* (= *Meliolaceae*), Saccardo & Sydow (1899) in *Microthyriaceae* subfam. *Asterinoideae*, Hennings (1908) in *Microthyriaceae*, and Arnaud (1921) in *Parodiellinaceae* (= *Parodiopsidaceae*).

*Asteronia sweetiae* has globose, ostiolate ascomata that form on black mycelium, while asci form from the base of ascomata. These characters and lack of flattened thyriothecia indicate that this genus is not well placed in *Microthyriaceae*. A more suitable family is probably *Asterinaceae* or *Meliolaceae*. However, there are no sequence data for *Asteronia* in GenBank and fresh collections are needed in order to establish the phylogenetic relationship of this genus.

***Dictyoasterina*** Hansf., Proceedings of the Linnean Society of London  
159: 39, 1947.

FIG 2A–K

TYPE SPECIES: *Dictyoasterina conopharyngiae* Hansf., Proceedings of the Linnean Society of London 159: 39, 1947.

Epiphytic on the upper surface of leaves, appearing as blackened areas, which are rounded and shiny, scattered over the leaf, with superficial hyphae present, forming a black mycelia net (FIGS 2A, B). Superficial hyphae, black to brownish, parallel and anastomosing, with sparse, lateral hyphopodia, hyphopodia nearly circular, black-brown, 20.5–28  $\mu\text{m}$  (mean = 26  $\mu\text{m}$ ) (FIG 2D). Ascomata 38–100  $\mu\text{m}$  high, 90–220  $\mu\text{m}$  diam, superficial, gregarious, roughly globose, black, subcoriaceous or less carbonaceous, with a central ostiole (FIG 2C). Peridium 6–18  $\mu\text{m}$  wide comprised of three layers of cells, black-brown to pale brown, outer layer of black brown cells compressed, inner layer of isodiametric cells of textura epidermoidea (FIGS 2E, G). Hamathecium of dense, long pseudoparaphyses, 50–61  $\times$  1.5–3  $\mu\text{m}$  (mean = 55  $\times$  2.4  $\mu\text{m}$ ) unbranched (FIGS 2F, I). Asci 32–47.5  $\times$  8–13  $\mu\text{m}$  (mean = 35.9  $\times$  9.9  $\mu\text{m}$ ), 8-spored, bitunicate, fissitunicate, clavate to short cylindrical, with a short knob-like pedicel 1.7–3.2  $\mu\text{m}$ , and inconspicuous apical structure (FIG 2H). Ascospores 9.5–15  $\times$  3–6  $\mu\text{m}$  (mean = 11.8  $\times$  4.4  $\mu\text{m}$ ), biseriate to overlapping triseriate, oblong-ellipsoid to obovate, 1-septate, hyaline, strongly constricted at the septum, upper cell oval, wider and shorter than the cone-shaped lower cell, guttulate, smooth-walled (FIGS 2J, K).





FIG. 2. *Dictyoasterina conopharyngiae* (from holotype). A. Appearance of fungus on surface of leaf. B. Appearance of colony and ascomata on the host surface. C. Squash mount of ascoma. D. Hyphopodia. E. Section of ascoma. Note the peridium which comprises several layers of cells. F. Asci and paraphyses. G. Peridium. H. Asci mounted in Melzer's reagent. I. Paraphyses. J, K. Ascospores becoming red when mounted in Congo red and yellow to light brown in Melzer's reagent. Scale bars: B = 200µm, C = 20 µm, D–K = 10 µm.

SPECIMEN EXAMINED: UGANDA, Entebbe, on leaf of *Conopharyngia holstii* (*Apocynaceae*), December 1945, C.G. Hansford, (IMI5298, holotype).

*Dictyoasterina* was described as a monotypic genus represented by *D. conopharyngiae* and was placed into the family *Asterinaceae* (Hansford 1947). In the latest Myconet, *Dictyoasterina* is removed into the family *Microthyriaceae* (Lumbsch & Huhndorf 2007).

*Dictyoasterina conopharyngiae* has superficial mycelium and clearly ostiolate, globose ascomata; these morphological characters make it distinct





FIG.3. *Helminthopeltis almeidaeana* (from holotype). A. Appearance of ascomata on the host surface. B, C. Squash mount of ascoma. D–F, L. Section of ascomata. Note peridium. G, I, K, M. Unitunicate asci. H, J. Ascospores. Scale bars: A = 500  $\mu\text{m}$ , B = 20  $\mu\text{m}$ , C–M = 10  $\mu\text{m}$ .

from *Microthyriaceae*. We suggest that *Dictyoasterina* be excluded from *Microthyriaceae* and transferred to a more suitable family, such as the

*Asterinaceae*, although the presence of pseudoparaphyses should be considered. As there are no sequences for this genus in GenBank, fresh collections and DNA sequence analyses are still needed to establish taxonomic placement.

***Helminthopeltis*** Sousa da Câmara, *Agronomia Lusitana* 12: 102, 1950. FIG 3A–M

TYPE SPECIES: *Helminthopeltis almeidaeana* Sousa da Câmara, *Agronomia Lusitana* 12: 102, 1950.

Necrotrophic or biotrophic on the surface of leaves of conifers, forming black oval spots. Superficial mycelium absent. *Ascomata* 140–205 µm long × 75–110 µm wide, about 100–150 µm high, solitary or grouped in pairs, scattered, superficial, clypeate, oblong or elongated, with a longitudinal cleft-like opening, subcoriaceous, black to brown (FIGS 3A, B, C). Peridium 50–72 µm thick at the apex, 25–35 µm thick at base, two-layered, composed of hyaline pseudoparenchymatous cells and an inner layer of isodiametric cells of *textura angularis* (FIGS 3D–F, L). Hamathecium sparse or absent. Asci 60–117 × 10–18 µm (mean = 77 × 15.2 µm), at least 8-spored (or more than 8-spored), not fissitunicate, cylindrical to oblong, thin-walled, parallel arrangement (FIGS 3G, I, K, M). Ascospores 62.5–83 × 2–3.5 µm (mean = 70.9 × 2.8 µm), parallel seriate, broad filiform to wire-like, hyaline, aseptate, wall smooth (FIGS 3H, J).

SPECIMENS EXAMINED: PORTUGAL, Minho Serra do Gerez (Pico Borrageiro), on leaf of *Juniperus communis* (*Cupressaceae*), 3 July 1948, M. de Sousa da Câmara (LISE 50024, holotype).

Sousa da Câmara (1950) erected *Helminthopeltis* as a monotypic genus for *H. almeidaeana*, which is found only in Europe (Kirk et al. 2008). The 2007 Outline of *Ascomycota* (Lumbsch & Huhndorf 2007) places *Helminthopeltis* into *Microthyriaceae*. We suggest that *H. almeidaeana* is better placed in *Lophodermium* (*Rhytismataceae*) but do not transfer the species here as it has probably already been described in this large genus under another name.

***Hidakaea*** I. Hino & Katum., *Bulletin of the Faculty of Agriculture, Yamaguchi University* 6: 38, 1955. FIG 4A–K 5A–D

TYPE SPECIES: *Hidakaea tumidula* I. Hino & Katum., *Bulletin of the Faculty of Agriculture, Yamaguchi University* 6: 38, 1955.

Saprobic or parasitic on stems of bamboo, causing black to brown spots. *Ascomata* 260–270 µm diam. 120–160 µm high, solitary or gregarious, flattened against the host surface, subglobose, in section hemispherical, brown, subcoriaceous but membranous at the base, smooth from above, unilocular, with a central ostiole (FIGS 4A, B). Ostiole 6.0–6.5 µm. Peridium 41–63 µm wide, brown-black at the sides, other parts light brown, in the sagittal section, comprising a few layers of cells, outer cells appear pseudoparenchymatous and cell wall very thin, on the base cells isodiametric, black-brown (FIGS 4C,



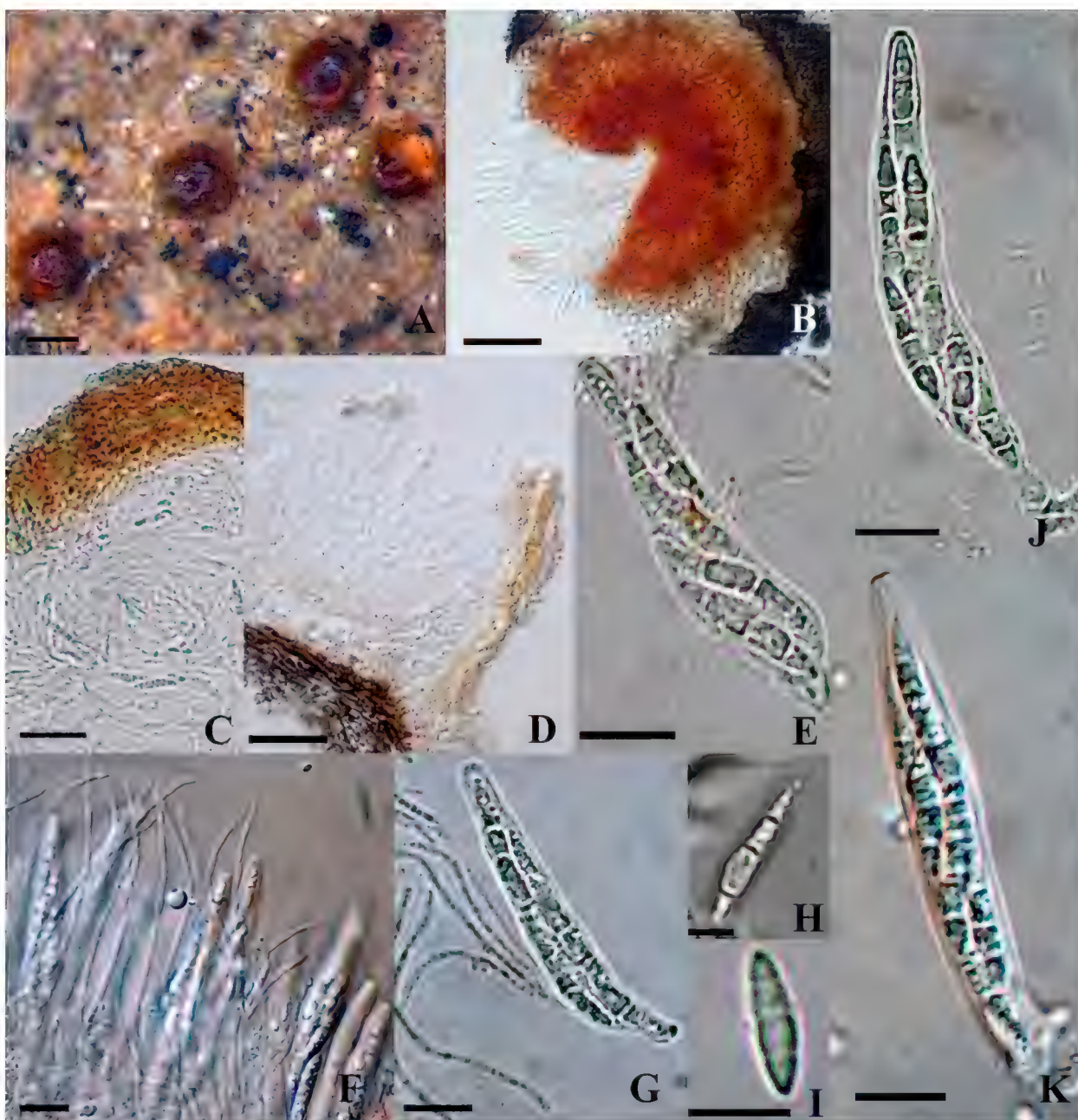


FIG. 4. *Hidakaea tumidula* (from holotype). A. Appearance of ascomata on the host surface. B. Squash mount of ascoma. C, D. Section of ascoma. Note the peridium. E, J, K. Asci. Note the pedicel and the apical ring structure. F, G. Paraphyses which are longer than asci. H, I. Ascospores. Note the three septa and inconspicuous sheath. Scale bars: A = 200  $\mu\text{m}$ ; B, C, D = 20  $\mu\text{m}$ ; E–G, J, K = 10  $\mu\text{m}$ ; H, I = 5  $\mu\text{m}$ .

D). Hamathecium comprising paraphyses,  $70\text{--}105 \times 1\text{--}2 \mu\text{m}$ , embedded in mucilage, and longer than asci (FIGS 4F, G). Asci  $71\text{--}82 \times 7\text{--}10 \mu\text{m}$ , 8-spored, unitunicate, cylindrical-clavate or oblong, with a short pedicel  $6 \times 4 \mu\text{m}$ , apically rounded with inconspicuous apical structure (FIGS 4E, J, K). Ascospores  $16\text{--}20.5 \times 2.5\text{--}4 \mu\text{m}$ , 2-seriate, fusiform, tapering gradually at one or both ends, or pointed, hyaline or pale, 3-septate, slightly constricted at septa, smooth-walled, some with sheath (FIGS 4H, I).

SPECIMEN EXAMINED: JAPAN, Sagami Province, Qoyama, on dead stems of *Pleiblastus vaginatus* (*Poaceae*, *Bambusoideae*), September 1952, Hino and Katumoto (YAM 20296, holotype).

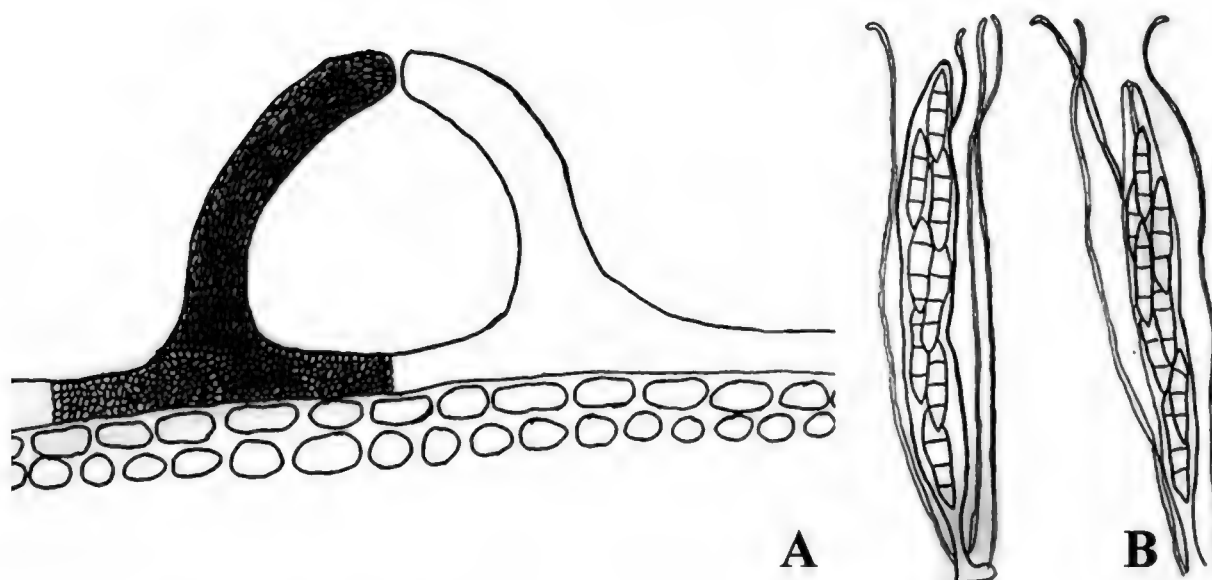


FIG.5. *Hidakaea tumidula* (line drawing from holotype). A. Ascoma. B. Asci.

Hino & Katumoto (1955) established *Hidakaea* as a monotypic genus with *H. tumidula* as the type species and suggested that it should be placed into *Microthyriaceae* based on ascomata with a scutate structure, superficial subglobose ostiolate ascomata, and bitunicate, clavate or cylindrical-clavate asci. The scutate structure and bitunicate asci described in the prologue were not observed in the holotype. The unitunicate ascus of *Hidakaea* does not belong in *Dothideales* and *Microthyriaceae*. The species should be placed the *Sordariomycetes* where it may have affinities with the *Chaetosphaeriales* or *Hypocreales*. The brown colour of the ascomata is typical of species in the *Hypocreales*, and the asci and ascospores are also characteristic of this order. We therefore suggest that *Hidakaea* belongs in *Hypocreales* incertae sedis.

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We express our deep thanks to Drs Eric McKenzie and Cai Lei for reviewing the manuscript. The Research Institute of Resource Insects, Chinese Academy of Forestry provided financial support to Wu Haixia Master's study. Funds for research were provided by the Grant for Essential Scientific Research of National Non-profit Institute (no. CAFYBB2007002). The curators of the following herbaria are thanked for providing materials on loan for this study: K, IMI, LISE, and YAM. We also thank Professor Xiaoming Chen for much help.

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## MYCOTAXON

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**A new species of non-lichenized genus *Stictis*  
(*Ostropales*, *Lecanoromycetes*) from India**

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**Abstract** — *Stictis subbrachyspora* is described as new species from India. The new species is characterized by a non-lichenized habit; round, effigurate, chroodiscoid apothecia with white pruinose, lacerate margins, and transversely 19–32 loculate small ascospores measuring  $45\text{--}55 \times 4\text{--}5 \mu\text{m}$ . *Stictis himalayanus* is published as a new combination based on *Chroodiscus himalayanus*.

**Key words** — *Stictidaceae*, corticolous, *Ascomycota*, *Conotrema*

**Introduction**

The family *Stictidaceae* (*Ostropales*, *Lecanoromycetes*, *Ascomycota*) accommodates a large group of lichenized and non-lichenized fungi. The species of the genus *Stictis* Pers. are perhaps the best known example of individuals representing the same fungal species having the ability to live either in a lichenized state (with algal symbionts) or as saprotrophs (non-lichenized), depending on the substrate (Wedin et al. 2004).

Sherwood (1977a,b, 1979) provided a comprehensive account of c. 65 species and separated *Stictis* from allied genera based on the orbicular fruiting body opening by pores, periphysoids in an apothecial margin that extends down the whole length, a hymenium that splits away from the margin when dry, a thick crystalline layer in the ascoma margin and a non-parasitic niche. The ascospores exhibit variation in shape (cylindrical or filiform), size, and septation (with 3–300 septa). *Stictis* was segregated from the closely related *Conotrema* based on the lichenized living strategy and scattered crystals in apothecial margin of the latter (Gilenstam 1969, Sherwood 1977a). A phylogenetic account by Wedin et al. (2006) suggests that *Stictis* is paraphyletic and congeneric with

*Conotrema*. The other closely related genera — *Schizoxylon* and *Carestiella* — differ from *Stictis* in lacking a periphysoidal layer and having disintegrated spores (Wedin et al. 2005).

Seven *Stictis* taxa have been reported previously from India: *S. bengalensis* U.P. Singh & Pavgi, *S. indica* Tilak & Nanir, *S. kamatii* Tilak & S.B. Kale, *S. lantanae* Tilak & Nanir, *S. marathwadensis* Tilak & S.B. Kale, *S. stellata* subsp. *intermedia* (Speg.) M.P. Sharma & R. Sharma, and *S. tilakii* S.B. Kale & S.V.S. Kale (Kale & Kale 1970, Sharma & Sharma 1983, Singh & Pavgi 1966, Tilak & Kale 1969, 1970; Tilak & Nanir 1975). Some of these are no longer considered to belong in *Stictis*, and others are of dubious application. Sherwood (1977a) synonymized *S. bengalensis* under *S. radiata* (L.) Pers. subsp. *radiata* and recombined *S. lantanae* as *Schizoxylon lantanae* (Tilak & Nanir) Sherwood, placing *S. indica* in synonymy. Sherwood (1977a) was unable to obtain material of *S. kamatii*, *S. marathwadensis*, and *S. tilakii* for study; on the basis of their protologues, she considered that *S. marathwadensis* was probably a *Stictis* sp. distinct from *S. radiata* but was unable to suggest better taxonomic placements for *S. kamatii* and *S. tilakii*.

The present paper reveals the existence of a non-lichenized corticolous species from India that fits well in the genus *Stictis* and is described here as new to science.

## Materials and methods

The material (preserved in LWG, the National Botanical Research Institute lichen herbarium) was examined morphologically, anatomically, and chemically. Thin hand-cut sections of apothecia and thallus were mounted in plain water, cotton blue, 5% KOH, and iodine solution and observed under a compound microscope. Chemical spot tests and TLC methods follow methods by Orange et al. (2001). The minimum and maximum measurements of ascospore and other anatomical features are based on the examination of at least five different mature ascomata.

## Taxonomic description

*Stictis subbrachyspora* S. Joshi & Upreti, sp. nov.

FIGURE 1

MYCOBANK MB518076

*Thallus corticolus. Ascocarpi primum immerse, erumpescentes, profunde cupulati, 0.5–0.6 mm diam., margin, albo, lacerato. Margo in sectione 60–120(–125) µm crassus. Exipulum proprium brunneum, 30–65 µm crassum; stratum crystallinum 25–50 µm crassum. Periphysioidea ramose, 15–25 µm. Paraphyses filiformes, ramosae, 1–1.5 µm. Asci 8-spori, 80–90(–100)×5–6 µm crassum, I+ carulescens. Sporae 45–55 × 4–5 µm, involutae, I+ aurantiaco-lutescentes.*

TYPUS: INDIA, West Bengal, Jalpaiguri district, Jaigaon, on bark on a river bank, 07.04.1987, D.K. Upreti & M. Ranjan 201675 (LWG-holotype).

A non-lichenized fungus, corticolous, forming a hyaline epiphloeodal hyphal felt or thallus, 20–50  $\mu\text{m}$  thick. Ascomata, urceolate, solitary, sometimes aggregated in two, at first immersed, opening by a pore, becoming erumpent and finally nearly superficial, 0.5–0.6 mm in diam., round, chroodiscoid; disc brownish to flesh coloured, densely pruinose, splitting away from the margin, up to 0.4 mm in diam., deeply immersed; margin radiate, effigurate, lacerate, 5–6 lobed, white-pruinose, eroded in older apothecia, 60–120(–145)  $\mu\text{m}$  thick in cross section, hyaline to darken in older apothecia, sometimes layered, entirely encrusted in crystals.

Outer exciple layer 40–80  $\mu\text{m}$  thick; inner exciple layer brown, 30–65  $\mu\text{m}$  thick, branched periphysoids present, forming the innermost layer of the margin, 15–25  $\mu\text{m}$  long, separated from the outer wall by crystals (of 20–40  $\mu\text{m}$  in size); crystals forming a dense layer along the inner margin of apothecia, 25–50  $\mu\text{m}$  thick. Epihymenium indistinct, granular, hyaline to slightly brownish, usually covered by 20–40  $\mu\text{m}$  high crystalline layer, hymenium hyaline, inspersed, separated from the margin in dry condition, 100–135(–200)  $\mu\text{m}$  high, I+ golden yellow to wine-red; sub-hymenium, 30–50(–260)  $\mu\text{m}$  high, hyaline to darken in older apothecia, I+ blue. Paraphyses filiform, branched, with thickened apical cell, dense, conglutinate, 1.0–1.5  $\mu\text{m}$  wide, I+ blue in epihymenial region. Ascus 8-spored, cylindrical, bitunicate, 80–90(–200)  $\times$  5–6  $\mu\text{m}$ , I+ blue; ascospores cylindrical to fusiform, hyaline, transversely septate, sheathed, 19–32 loculate, locules broader than longer, 45–55  $\times$  4–5  $\mu\text{m}$ , golden yellow in Iodine solution.

**CHEMISTRY:** Thallus K+ reddish, PD–, C–; no lichen substance in TLC (Solvent system A).

**DISTRIBUTION AND ECOLOGY:** At present the new species is known only from the northern and eastern states of India, where it is found growing luxuriantly on tree bark in tropical moist deciduous forest at 140–900 m altitudes.

*ADDITIONAL SPECIMEN EXAMINED:* INDIA, Uttarakhand, Jim Corbett Tiger Reserve, Dugadda, on tree bark, 03 Dec. 1999, D. K. Upreti 217467 (LWG).

**REMARKS:** *Stictis subbrachyspora* is characterized by non-lichenized thalli, round erumpent, chroodiscoid apothecia with radiate, lacerate, pruinose margins, flesh coloured deeply immersed discs, branched paraphyses, periphysoids in the innermost layer, a crystalline layer between excipulum and periphysoids, brown inner exciple, and relatively small 8-spored asci and ascospores measuring 80–90(–100)  $\times$  5–6  $\mu\text{m}$  and 45–55  $\times$  4–5  $\mu\text{m}$  respectively.

*Stictis brachyspora* Sacc. & Berl. is similar to *S. subbrachyspora* in its white pruinose apothecial disc, thick crystalline layer separating periphysoids from outer wall, I+ blue paraphyses at epihymenial region, and an amyloid hymenium that splits away from the margin when dry; it differs in having broadly open immersed apothecia that do not become erumpent, larger ascospores (65–90



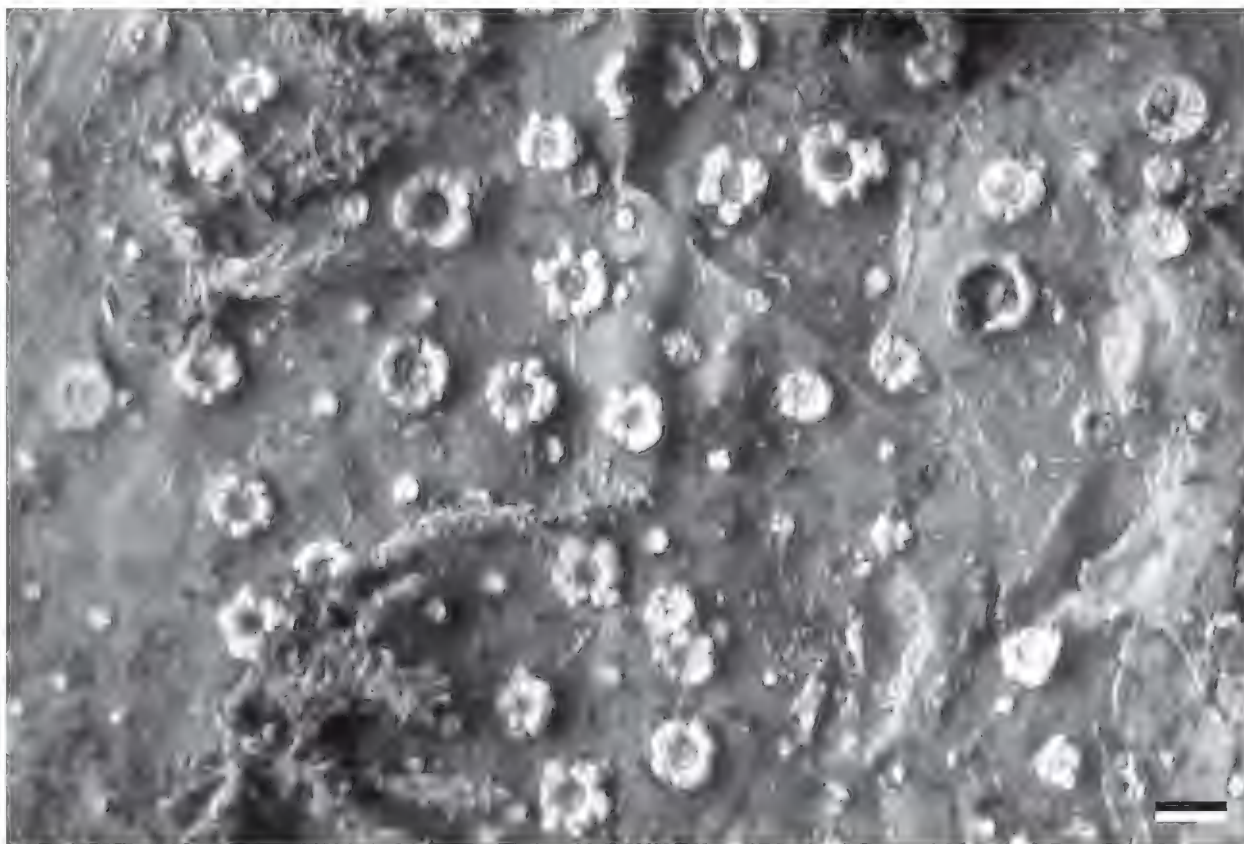


FIGURE 1. Thallus and apothecial morphology of *Stictis subbrachyspora* (Holotype).  
Scale: 0.5 mm.

$\times 3.5\text{--}4.5\text{ }\mu\text{m}$ ), and an apothecial margin that is  $40\text{ }\mu\text{m}$  thick and colourless throughout in cross section.

*Stictis friabilis* (W. Phillips & Plowr.) Sacc. & Traverso resembles the new species in its  $0.3\text{--}0.6\text{ mm}$ , erumpent, nearly superficial apothecia with deeply fissured pruinose margins, branched propoloid paraphyses, and small ascospores ( $55\text{--}70 \times 2.5\text{--}3.5\text{ }\mu\text{m}$ ). However, *S. friabilis* differs in having I-paraphyses in epihymenial region, a distinctly reddish apothecial disc with a fleshy yellow-pruinose margin, colourless proper exciple, and unsheathed ascospores (the sheathed ascospore character is not cited in the description by Sherwood, 1977a).

Other closely related taxa *S. lupini* W. Phillips & Harkn. and *S. dumontii* Sherwood, with small ascospores of  $45\text{--}60 \times 3\text{--}3.5\text{ }\mu\text{m}$  and  $55\text{--}65 \times 3\text{--}3.5\text{ }\mu\text{m}$  respectively, differ from *S. subbrachyspora* in having immersed apothecia. Further, *S. lupini* differs in having an entire apothecial margin, unbranched periphysoids, simple paraphyses, and indistinct exciple while *S. dumontii* differs in having 4-spored asci.

*Stictis radiata* subsp. *radiata* is comparable to new species in having a lacerate, white-pruinose, apothecial margin, deeply immersed disc, I+ blue subhymenial region, and irregularly branched paraphyses; however, the larger asci ( $120\text{--}250 \times 5\text{--}8\text{ }\mu\text{m}$ ) and unsheathed larger ascospores differentiate it from *S. subbrachyspora*.

*Stictis marathwadensis*, considered a good representative of the genus, is close to *S. subbrachyspora* in having round apothecia with lobed margins but differs in having white to black apothecia and acicular ascospores that are almost as long as the asci ( $220\text{--}285 \times 0.6\text{--}1.6 \mu\text{m}$ ).

The genus *Stictis* has a worldwide distribution but most species are common in coastal areas and moist humid cloud forests of tropical countries.

An Indian specimen previously placed in the otremataceous group due to uncertainty in the delimitation of *Ostropales* has been reevaluated and is now transferred to *Stictis* as follows:

***Stictis himalayanus*** (Nayaka & Upreti) S. Joshi & Upreti, **comb. nov.**

MYCOBANK MB518077

BASIONYM: *Chroodiscus himalayanus* Nayaka & Upreti, Mycotaxon 98: 247. 2006.

The taxon is characterized by chroodiscoid apothecia with prominent white exfoliating margins, hyaline proper exciples densely interspersed with calcium oxalate crystals, distinct periphysoids, and acicular transversely septate golden yellow I+ ascospores measuring  $40\text{--}78(-85) \times 3\text{--}5 \mu\text{m}$ . It is similar to *Stictis lupini* and *S. brachyspora* in having simple paraphyses and small ascospores measuring  $45\text{--}60 \times 3\text{--}3.5 \mu\text{m}$  and  $65\text{--}90 \times 3.5\text{--}4.0 \mu\text{m}$  respectively but differs in having erumpent apothecia, orange-brown discs, radially fissured apothecial margins, and 2–3 spored asci. The species is restricted to the Himalayas and was found growing on trees in Great Himalayan National Park, Himachal Pradesh, at an altitude of 2200 m.

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## MYCOTAXON

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***Perisporiopsis lateritia*, a new species on decaying leaves of *Hevea* spp. from the Amazon basin in Peru**

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**Abstract** — The genus *Perisporiopsis* (Ascomycota, Dothideomycetes, Parodiopsidaceae) occurs on the underside of decaying leaves, mostly in tropical regions. A new species of *Perisporiopsis*, *P. lateritia*, is described that can be distinguished from other species in the genus by a combination of teleomorph and anamorph characteristics, such as ascospore size, size and shape of microconidia and macroconidia of the *Septoidium* anamorph, and the plant host (*Hevea*). This species is known only from the Peruvian Amazon.

**Key words** — leaf litter fungi, loculoascomycetes, systematics, taxonomy

**Introduction**

The plant genus *Hevea* (Euphorbiaceae) is known for the ability to produce latex that is processed to obtain natural rubber. The best-known species for production of natural rubber is *Hevea brasiliensis* (Willd. ex A. Juss.) Müll. Arg. Other species in the genus include *H. benthamiana* Müll. Arg., *H. guianensis* Aubl., *H. nitida* Müll. Arg., and *H. pauciflora* (Spruce ex Benth.) Müll. Arg., as well as others that are rare (Schultes 1956). Although *Hevea* includes economically important species, the fungi associated with these hosts have not been explored (Araujo et al. 2004, Gazis & Chaverri 2010). As part of a study to characterize endophytic and ex planta fungi, e.g. saprophytes, of wild trees of *H. brasiliensis* and *H. guianensis*, ascomata of an unidentified species of *Perisporiopsis* Henn. (Ascomycota, Dothideomycetes, Parodiopsidaceae) were collected from decaying leaves in two locations in the Peruvian Amazon. Based on morphological data, this unidentified ascomycete is described here as a new species. A diagnostic sequence of the internal transcribed spacer region of the nuclear ribosomal DNA (ITS) has been deposited in Genbank.



## Materials & methods

### Source of specimens

Decaying leaves were collected near the base of wild *Hevea brasiliensis* and *H. guianensis* trees in old growth forests in two sites in the Peruvian Amazon, i.e. Los Amigos and Tambopata (Dept. Madre de Dios, Prov. Manu and Tambopata, respectively). Two specimens (P.C. 811 and P.C. 987) included ascomata of this unusual fungus. Ascospore germination was attempted by isolating asci and ascospores onto BBL™ cornmeal-dextrose-agar (CMD), supplemented with antibiotics (Sigma-Aldrich streptomycin-neomycin-penicillin). Plates were incubated at 25°C with alternating 12 h light/12 h darkness. However, ascospores did not germinate after one week. Specimens are preserved in the U.S. National Fungus Collection (BPI).

### Morphological characterization

To observe internal and microscopic characteristics, the ascomata were rehydrated briefly in KOH, then supported by Tissue-Tek O.C.T. Compound 4583 (Miles Inc., Elkhart, Indiana, U.S.A.), and sectioned with a freezing microtome at a thickness of ca. 15 µm. Characteristics of asci and ascospores were observed by rehydrating the ascomata in 3% KOH, removing part of the centrum with a fine glass needle, and placing it on a glass slide. Characteristics of the anamorph on the natural substrata were also observed. Measurements of continuous characters such as length and width were made using Scion Image software beta version 4.0.2 (Scion Corporation, Frederick, Maryland, U.S.A.). Continuous measurements are reported as the extremes (maximum and minimum) in brackets separated by the 95% confidence interval. Color terminology is from Rayner (1970).

### Source of ITS sequence

To obtain a representative ITS sequence, DNA was extracted from the ascomata of P.C. 811 by removing centrum contents with a fine glass needle and placing them in the bead-beating microtubes of the PowerPlant™ DNA isolation kit (MO BIO Laboratories, Inc., Carlsbad, California, U.S.A.). The primers used for ITS were ITS 5 and ITS 4 (White et al. 1990). PCR reactions were run in an Eppendorf Mastercycler EP using the parameters described in Gazis & Chaverri (2010). PCR products were cleaned using ExoSAP-IT® (USB Corporation, Cleveland, Ohio, U.S.A.) following the manufacturers instructions. Clean PCR products were sequenced at the DNA Sequencing Facility (Center for Agricultural Biotechnology, University of Maryland, College Park, Maryland, U.S.A.). Sequences were assembled and edited with Sequencher 4.9 (Gene Codes, Madison, Wisconsin, U.S.A.). The ITS sequence was deposited in Genbank as accession number FJ884129.

## Taxonomy

***Perisporiopsis lateritia*** P. Chaverri & Gazis, sp. nov.

PLATE A–H

MYCOBANK — MB518067

*Perisporiopsis melioloides similis*. Ascospores (65.0–)66.0–75.5(–78.0) × 18.0–21.5(–23.0) µm. Septoidium macroconidia ovoideus fusiformes ad cymbiformes, (59.0–)61.5–69.0 (–80.0) × (15.7–)16.5–18.0(–19.3) µm, longitudo/crassitudo 3.7–3.8(–4.2). Microconidia

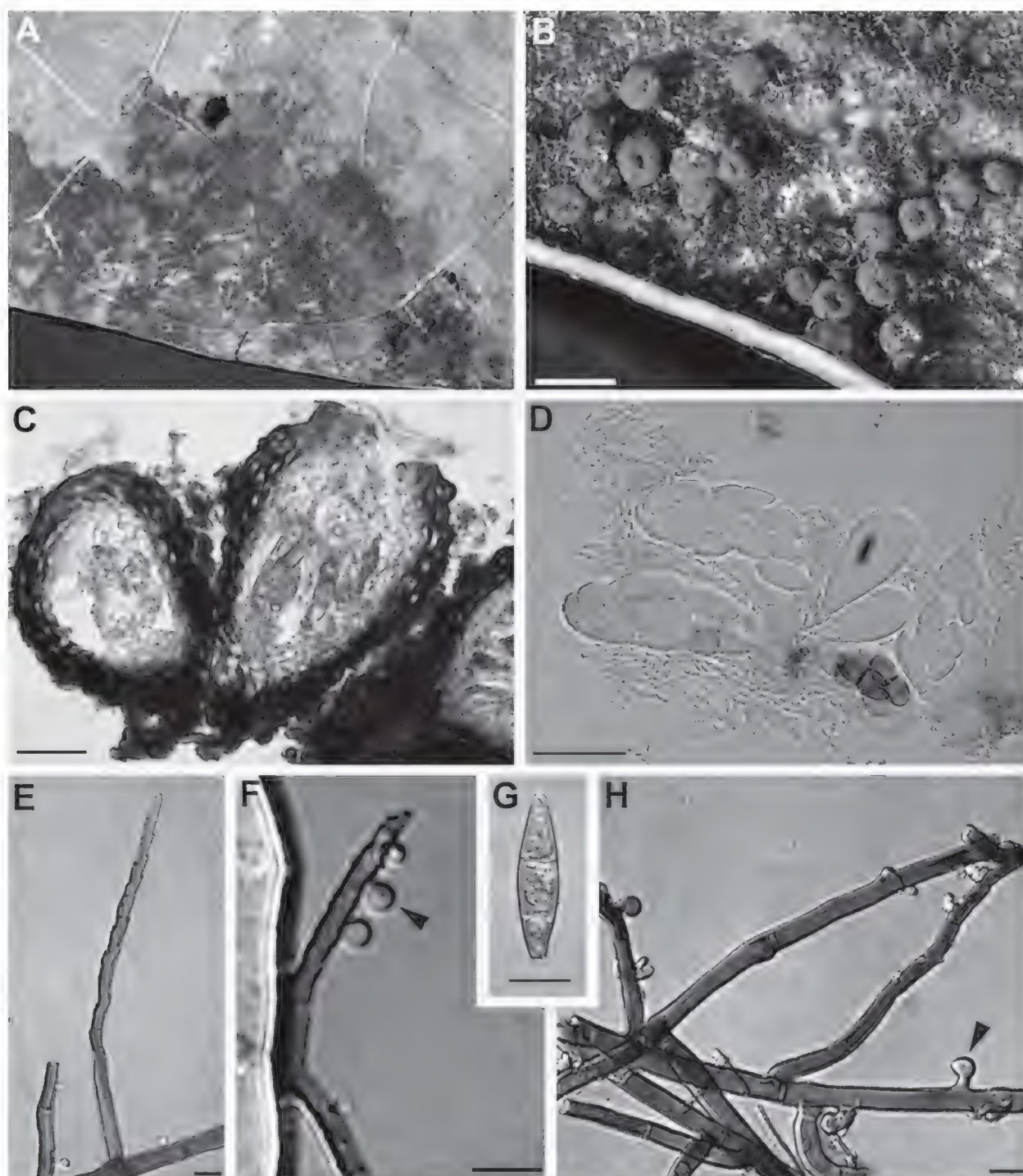


PLATE. *Perisporiopsis lateritia* Holotype P.C. 811 = BPI 880185. A, B. Ascomata and dark mycelium on the underside of leaves. C. Longitudinal section of ascomata. D. Asci and ascospores. E, F. Denticulate conidiophore of the microconidial anamorph. F. Arrow indicates globose microconidia. G. *Septoidium* macroconidium. H. Simple stomatopodia indicated by arrow.

Bars: B = 1 mm; C, D = 100  $\mu$ m; E–H = 10  $\mu$ m.

*globosae ad subglobosae*,  $4.5\text{--}5.5 \times 4.8\text{--}5.5 \mu\text{m}$ , *longitudo/crassitudo* 1.0–1.1. *Stomatopodii simplex*.

TYPE: 17 June 2007, coll. R. Gazis, H.C. Evans, P. Chaverri; (Holotype BPI 880185, P.C. 811) on underside of decaying leaves of *Hevea brasiliensis*, Picaflor Research Station, near Tambopata River, Prov. Tambopata: Dept. Madre de Dios, Peru. GenBank accession number FJ884129.

ETYMOLOGY: The name is Latin for brick red, in reference to the color of the ascomata.

TELEOMORPH – Mycelium superficial, hypophyllous, extensive, appearing black, anastomosing to form a close network, almost subiculum-like, with simple, knob-shaped stomatopodia. Ascomata superficial on mycelium, aggregated, associated with a hyphomycetous dematiaceous anamorph (i.e. *Septoidium*). Ascomata dark brown to black, almost completely covered with a sienna to brick tomentum, except near the apex where they appear black, subglobose to obovoid,  $300\text{--}310 \times 420\text{--}450 \mu\text{m}$  ( $n = 5$ ), non-ostiolate, irregularly dehiscent at apex; ascomatal wall composed of one region of 2–3 layers of thick-walled cells, textura angularis. Asci few, generally less than 5,  $200\text{--}220 \times 80\text{--}90 \mu\text{m}$  ( $n = 10$ ), obovoid, sessile to short stalked, somewhat thickened at apex, eight-spored. Ascospores 1-septate, strongly constricted at septum, initially hyaline, later pale brown or fawn, smooth to slightly spinulose, broadly fusiform to ovoid, somewhat inequilateral, with apical cell slightly larger than basal cell,  $(65.0\text{--})66.0\text{--}75.5(-78.0) \times 18.0\text{--}21.5(-23.0) \mu\text{m}$  (average =  $70.5 \times 20 \mu\text{m}$ ,  $n = 30$ ).

ANAMORPH – Both macro- and microconidia of the hyphomycetous anamorph observed on natural substrata. For the macroconidial anamorph (i.e. *Septoidium*) no conidiogenous cells observed. Macroconidia ovoid, fusiform to cymbiform, truncate at base, smooth, pale brown, sometimes with tinges of pale grayish rose, 2-septate,  $(59.0\text{--})61.5\text{--}69.0(-80.0) \times (15.7\text{--})16.5\text{--}18.0(-19.3) \mu\text{m}$  (average =  $65.2 \times 17.2 \mu\text{m}$ ,  $n = 10$ ), length/width ratio 3.7–3.8(–4.2) (average = 3.8,  $n = 30$ ). Microconidial anamorph with erect conidiophores, brown near base, pale brown almost hyaline near tip, simple, not branching, septate, with scattered denticles on upper part; conidiogenous cells polyblastic, sympodial, with small denticles; microconidia borne on denticles, globose to subglobose, unicellular, almost hyaline, sometimes apiculate at base,  $4.5\text{--}5.5 \times 4.8\text{--}5.5 \mu\text{m}$  (average =  $5 \times 5.2 \mu\text{m}$ ), length/width ratio 1.0–1.1 (average = 1.0,  $n = 8$ ).

HABITAT – On the underside of decaying *Hevea* spp. leaves in old growth forests. Known only from Peru.

ADDITIONAL SPECIMEN EXAMINED: PERU. DEPT. MADRE DE DIOS: PROV. MANU, LOS AMIGOS RESEARCH STATION, NEAR LOS AMIGOS RIVER, on underside of decaying leaves of *Hevea guianensis*, July 2007, coll. R. Gazis BPI 880186 (= P.C. 987).

NOTES – *Perisporiopsis* includes 19 species, all of them occurring on decaying leaves in tropical regions; most are described in Sivanesan (1984). A manuscript under review (Chaverri & Gazis) shows that *Perisporiopsis* is also a common endophyte and soil inhabitant. *Perisporiopsis lateritia* is most similar to *P. melioloides* (Berk. & M.A. Curtis) Arx in having a reddish tomentum covering the ascomata and relatively large ascospores. *Perisporiopsis melioloides* has conidia that are significantly wider than those of *P. lateritia*. In addition, the stomatopodia of *P. melioloides* are lobed while in *P. lateritia* they are simple. Other species with a reddish covering on the ascomata are *P. brasiliensis*



(Bat. & Nascim.) Arx, *P. cecropiae* (R.E.D. Baker) Arx, *P. fusispora* (Pat.) Arx, *P. kwangensis* (Henn.) Arx, and *P. megalospora* (Sacc. & Berl.) Arx. Most of these species have smaller ascospores than *P. lateritia*, and *P. fusispora* has multiseptate, fusiform ascospores. In addition, *P. brasiliensis* has *Septoidium* macroconidia that are generally 3-septate, *P. cecropiae* has macroconidia generally 1-septate, *P. fusispora* has 3-septate macroconidia, *P. kwangensis* has smaller microconidia, and *P. megalospora* has lobed stomatopodia and larger microconidia than *P. lateritia*.

Among species of *Perisporiopsis*, host preferences seem to exist, i.e. most species are from plants of close taxonomic affinity (Sivanesan 1984). For example, *Perisporiopsis brachystegiae* (Henn.) Arx and *P. fusispora* are known only from legumes in Africa and Tropical America, respectively. *Perisporiopsis megalospora* is known from various genera in the *Malpighiales* such as *Banisteriopsis*, *Hiraea*, *Mascagnia*, and *Tetrapteris*; and *P. melioloides* from the *Myrtaceae*. Only two other species have been found on *Euphorbiaceae*, namely *P. hurae* (R.E.D. Baker & Dale) Arx [“*urae*”] and *P. kwangensis*; these two species are morphologically distinct from *P. lateritia*.

All species of *Perisporiopsis*, except *P. lantanae* (F. Stevens) R.W. Barreto, have *Septoidium* macro- and microconidial anamorphs. In Barreto et al. (1995), *P. lantanae* is described as having a pycnidial anamorph, more typical of a *Leptosphaeria*. In addition, the ascospores illustrated in Barreto et al. (1995), resemble *Leptosphaeria*. Therefore, it is likely that this species may not belong in *Perisporiopsis*.

Whether the phenotypic characteristics used to separate species of *Perisporiopsis*, i.e. ascospores, conidia, and host, have phylogenetic significance remains unknown. This genus has not been included in phylogenetic studies of the *Dothideomycetes* (Schoch et al. 2009). Its relationship with other genera in the *Parodiopsidaceae* is unclear. Given the small ascomata, few asci, lack of interthecial elements and occurrence on leaves, one would suspect a relationship with the *Mycosphaerellaceae* sensu lato. However, in a recently submitted manuscript by Chaverri & Gazis, phylogenetic analyses of nuclear ribosomal DNA suggest a close relationship with *Leptosphaeriaceae* and *Phaeosphaeriaceae*.

**Key to species of *Perisporiopsis***

Modified from Sivanesan (1984)

- 1. Ascospores always one-septate, conidia transversely multiseptate or staurosporous .....2
- 1. Ascospores with one or more septa, conidia transversely multiseptate ..... 9
- 2. Conidia staurosporous .....3
- 2. Conidia straight. ....4



3. Ascospores 25–33 x 7–12  $\mu\text{m}$ , conidia 50–100 x 10–16  $\mu\text{m}$ , on *Lophira*  
(*Ochnaceae*) ..... *P. lophirae*
3. Ascospores 30–52 x 10–15  $\mu\text{m}$ , conidia 50–140 x 28–56  $\mu\text{m}$ , on legumes  
..... *P. brachystegiae*
4. Conidia 1–2-septate .....5
4. Conidia 2–3-septate .....8
5. Ascomata in shades of orange or red, not brown or black .....6
5. Ascomata brown or black. ....7
6. Ascospores 40–50 x 11–15  $\mu\text{m}$ , conidia 57–68 x 12–15  $\mu\text{m}$ , microconidia  
5–9 x 2–3  $\mu\text{m}$ , on *Alchornea*, *Pera*, *Sapium* and other *Euphorbiaceae*  
..... *P. kwangensis*
6. Ascospores 36–51 x 12–15  $\mu\text{m}$ , conidia 36–45 x 15–20  $\mu\text{m}$ , microconidia  
5–7.5 x 3–4  $\mu\text{m}$ , on *Cecropia* (*Cecropiaceae*) ..... *P. cecropiae*
6. Ascospores 40–55 x 12–16  $\mu\text{m}$ , conidia 60–80 x 12–15  $\mu\text{m}$ , microconidia  
7–10 x 5–7  $\mu\text{m}$ , on *Banisteriopsis*, *Hiraea*, *Mascagnia*, and *Tetrapteris*,  
and other *Malpighiales* ..... *P. megalospora*
6. Ascospores 30–75 x 16–21  $\mu\text{m}$ , conidia 55–79 x 12–16  $\mu\text{m}$ , microconidia  
4–7 x 3–5  $\mu\text{m}$ , on *Myrtaceae* ..... *P. melioloides*
6. Ascospores 66–76 x 18–22  $\mu\text{m}$ , conidia 61–69 x 16–18  $\mu\text{m}$ , microconidia  
4.5–5.5 x 4.5–5.5  $\mu\text{m}$ , on *Hevea* (*Euphorbiaceae*) ..... *P. lateritia*
7. Ascospores 35–45 x 20–24  $\mu\text{m}$ , conidia 40–63 x 17–21  $\mu\text{m}$ , microconidia  
absent, on *Mauria* (*Anacardiaceae*) ..... *P. escharoides*
7. Ascospores 40–45 x 10–14  $\mu\text{m}$ , conidia 40–56 x 18–20  $\mu\text{m}$ , microconidia  
3–5 x 1.5–2.5  $\mu\text{m}$ , on *Buddleja* (*Scrophulariaceae*) ..... *P. torrendii*
7. Ascospores 36–52 x 13–20  $\mu\text{m}$ , conidia 60–65 x 18–20  $\mu\text{m}$ , microconidia  
9–12 x 8–10  $\mu\text{m}$ , on *Oryctanthus* (*Loranthaceae*) ..... *P. sydowii*
7. Ascospores 55–66 x 17–21  $\mu\text{m}$ , conidia 56–80 x 23–38  $\mu\text{m}$ , microconidia  
2.5–4 x 2–2.5  $\mu\text{m}$ , on *Hura* (*Euphorbiaceae*) ..... *P. hurae* ["urae"]
8. Ascospores 27–38 x 12–15  $\mu\text{m}$ , conidia 50–72 x 12–14  $\mu\text{m}$ , microconidia  
5–7.5 x 4–6.5  $\mu\text{m}$ , on *Tapirira* ..... *P. brasiliensis*
8. Ascospores 45–60 x 12–16  $\mu\text{m}$ , conidia 62–80 x 16–22  $\mu\text{m}$ , microconidia  
6–8 x 2–3  $\mu\text{m}$ , on *Clusia* (*Clusiaceae*) ..... *P. clusiae*
9. Ascospores 1–5-septate, 52–86 x 11–15  $\mu\text{m}$ , conidia 65–100 x 14–17  $\mu\text{m}$ ,  
on *Struthanthus* and other *Loranthaceae* ..... *P. struthanthi*
9. Ascospores 1–3-septate .....10
10. Ascospores 50–70 x 9–12  $\mu\text{m}$ , conidia 50–77 x 12–14,  
on legumes ..... *P. fusispora*
10. Ascospores 60–82 x 9–14  $\mu\text{m}$ , conidia 80–100 x 15–18  $\mu\text{m}$ ,  
on *Calophyllum* (*Clusiaceae*) ..... *P. portoricensis*

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**Two new species of *Phylloporia*  
(*Basidiomycota*, *Hymenochaetaceae*) from China**

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**Abstract** — The knowledge of *Phylloporia* in China is briefly summarized, and an identification key to the Chinese species in the genus is supplied. Two new species, *P. hainaniana* and *P. oblongospora*, are described and illustrated. The former species is characterized by its triquetrous pileus in section, relatively larger pores (4–6 per mm), and bigger, ellipsoid basidiospores ( $4.6\text{--}5.6 \times 3\text{--}3.6 \mu\text{m}$ ). *Phylloporia oblongospora* differs from other species in the genus by its homogeneous context, larger pores (2–4 per mm), and oblong ellipsoid basidiospores ( $4\text{--}4.8 \times 2\text{--}2.5 \mu\text{m}$ ).

**Key words** — basidiomycetes, polypore, taxonomy, wood-rotting fungi

**Introduction**

*Phylloporia* Murrill was defined for annual and monomitic species with duplex context and tiny coloured spores (Ryvarden 1991). However, based on the molecular and morphological study, some perennial and dimitic species were included in the genus, and they all form a monophyletic clade (Wagner & Ryvarden 2002). A modified definition on genus was proposed by Wagner &

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Ryvarden (2002), which includes annual to perennial, monomitic to dimitic species with duplex context and tiny coloured spores. Twelve species are accepted worldwide, most occurring in the tropics (Murrill 1904, Ryvarden & Johansen 1980, Wagner & Ryvarden 2002).

During the study on wood-decaying fungi from southern China, two species of *Phylloporia* could not be identified to any known species. They are described in the present paper as *Phylloporia hainaniana* and *P. oblongospora*. In addition, an identification key to the species of *Phylloporia* occurring in China is provided.

### Materials and methods

The studied specimens were deposited in herbaria as cited below. The microscopic procedure follows Cui et al. (2007). In presenting the variation in the size of the spores, 5% of measurements were excluded from each end of the range, and given in parentheses. In the text the following abbreviations are used: IKI = Melzer's reagent, IKI- = negative in Melzer's reagent, KOH = 5% potassium hydroxide, CB = Cotton Blue, CB+ = cyanophilous, CB- = acyanophilous, L = mean spore length (arithmetic average of all spores), W = mean spore width (arithmetic average of all spores), Q = variation in the L/W ratios between the specimens studied, n = number of spores measured from given number of specimens. Sections were studied at magnification up to  $\times 1000$  using a Nikon Eclipse E 80i microscope and phase contrast illumination. Drawings were made with the aid of a drawing tube. Special colour terms follow Petersen (1996) and Anonymous (1969).

### Taxonomy

*Phylloporia hainaniana* Y.C. Dai & B.K. Cui, sp. nov.

FIG. 1

MYCOBANK MB 513324

*Carpophorum annuum, pileatum, imbricatum; facies pororum bubalina vel mellea, pori rotundi vel angulati, 4–6 per mm. Systema hypharum monomiticum, hyphae septatae sine fibulis, hyphae contexti 3–8  $\mu$ m, hyphae tomenti 4–9  $\mu$ m, setae nullae; sporae flavidae, ellipsoideae, crassitunicatae, 4.6–5.6  $\times$  3–3.6  $\mu$ m.*

TYPE. — China. Hainan Province, Qiongzong County, Limushan Nature Reserve, on living angiosperm tree, 23.V.2008 Dai 9460 (holotype in IFP).

ETYMOLOGY — *hainaniana* (Lat.): refers to Hainan, the province name in China.

FRUITBODY — Basidiocarps annual, pileate, a few imbricate, broadly attached, soft corky and without odour or taste when fresh, becoming corky when dry; pileus triquetrous in section, projecting up to 0.7 cm, 1 cm broad and 10 mm thick at base. Pileal surface olivaceous buff when fresh, becoming fulvous when dry, azonate, tomentose; margin obtuse, buff yellowish. Poroid surface buff when fresh, becoming cinnamon buff when dry, more or less shining; margin

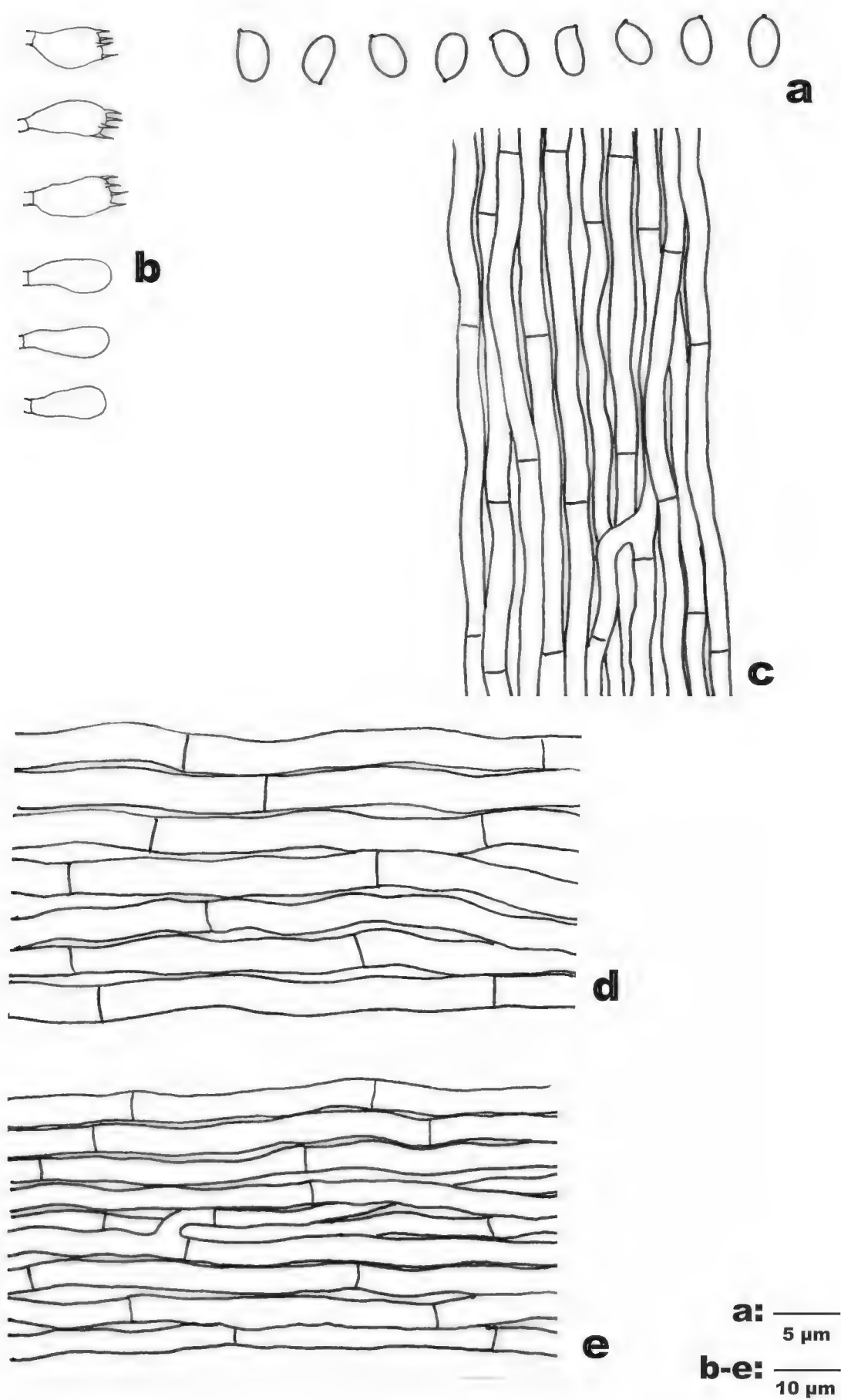


FIG. 1. Microscopic structures of *Phylloporia hainaniana* (drawn from the holotype).  
a: Basidiospores. b: Basidia and basidioles. c: Hyphae from tube trama.  
d: Hyphae from tomentum. e: Hyphae from context.

buff-yellow, narrow to almost lacking; pores circular or angular, 4–6 per mm, dissepiments thin, entire to slightly lacerate. Context cinnamon to fulvous, corky, up to 8 mm thick, duplex, a black line present, lower context hard corky, ca. 3 mm thick, upper tomentum soft corky, ca. 5 mm thick. Tubes cinnamon, slightly darker than pore surface, up to 2 mm long.

**HYPHAL STRUCTURE** — Hyphal system monomitic; all septa without clamp connections; tissue becoming bloody red but otherwise unchanged in KOH.

**CONTEXT** — Hyphae in the lower context pale yellowish brown, thin- to slightly thick-walled with a wide lumen, rarely branched, regularly arranged, 3–8  $\mu\text{m}$  in diam; hyphae of tomentum yellowish brown, thin- to slightly thick-walled with a wide lumen, rarely branched, frequently simple septate, straight, regularly arranged, moderately CB+, some collapsed, 4–9  $\mu\text{m}$  in diam; hyphae in the black zone dark brown, distinctly thick-walled with a narrow lumen, strongly agglutinate, winding and interwoven.

**TUBES** — Tramal hyphae hyaline to pale yellowish brown, thin-walled, occasionally branched, frequently simple septate, straight, parallel along the tubes, weakly or moderately CB+, 3–5  $\mu\text{m}$  in diam. Setae absent; basidia clavate, with four sterigmata and a simple septum at the base,  $13\text{--}23 \times 4\text{--}6 \mu\text{m}$ ; basidioles mostly pear-shaped, slightly smaller than basidia.

**SPORES** — Basidiospores ellipsoid, yellowish, fairly thick-walled, usually bearing a small guttule, more or less collapsed, IKI–, moderately CB+,  $(4.2\text{--})4.6\text{--}5.6 (-6) \times (2.8\text{--})3\text{--}3.6(-3.9) \mu\text{m}$ ,  $L = 5 \mu\text{m}$ ,  $W = 3.11 \mu\text{m}$ ,  $Q = 1.61$  ( $n = 30/1$ ).

**ADDITIONAL SPECIMEN (PARATYPE) EXAMINED** — China. Hainan Prov., Ledong County, Jianfengling Nature Reserve, angiosperm twig, 17.XI.2007 Cui 5160 (BJFC).

**REMARKS** — The pileate basidiocarps with a tomentum, a monomitic hyphal structure, ellipsoid, yellowish, and fairly thick-walled basidiospores, and a growth on living stem of shrub, make the species distinct in *Phylloporia*. The basidiospores of this species ( $4.6\text{--}5.6 \times 3\text{--}3.6 \mu\text{m}$ ) are the largest so far recorded for the genus, all other species having basidiospores less than 5  $\mu\text{m}$  in greatest dimension (Wagner & Ryvarden 2002).

*Phylloporia verae-crucis* (Berk. ex Sacc.) Ryvarden has slightly smaller basidiospores ( $4\text{--}4.5 \times 3\text{--}3.5 \mu\text{m}$ , Wagner & Ryvarden 2002); however, it sometimes has a laterally stipe, and its pores are distinctly smaller (7–9 per mm, Wagner & Ryvarden 2002). In addition, it lives on soil over buried wood, and occurs in South America (Wagner & Ryvarden 2002).

Following the identification key to the genus by Wagner & Ryvarden (2002), *Phylloporia hainaniana* would be close to *Phylloporia ampelina* (Bondartsev & Singer) Bondartsev, which has brittle and chalky basidiocarps and staining upper surface. In addition, its basidiospores are smaller ( $3.2\text{--}4 \times 2.5\text{--}2.8 \mu\text{m}$ ), and it grows on *Vitis* and is found so far in Central Asia (Bondartsev 1953).

***Phylloporia oblongospora* Y.C. Dai & H.S. Yuan, sp. nov.**

FIG. 2

MYCOBANK MB 513325

*Carpophorum annuum*, pileatum; facies pororum fulva, pori poroundi vel angulati, 2–4 per mm. Systema hypharum monomiticum, hyphae septatae sine fibulis, hyphae contexti 4–6  $\mu\text{m}$ , setae nullae; sporae flavidae, oblongo-ellipsoideae, crassitunicatae, 4–4.8  $\times$  2–2.5  $\mu\text{m}$ .

TYPE. — China. Guangxi Auto. Reg., Longzhou County, Nonggang Nature Reserve, on living branch of angiosperm tree, 14.VII.2007 Zhou 179 (holotype in IFP).

ETYMOLOGY — *oblongospora* (Lat.): refers to oblong ellipsoid basidiospores.

FRUITBODY — Basidiocarps annual, pileate, soft corky and without odour or taste when fresh, becoming corky to fragile when dry, pileus circular, projecting up to 2 cm, 3 cm broad and 5 mm thick at base. Pileal surface yellowish brown when dry, concentrically zonate, velutinate to smooth; margin sharp, concolorous to pileal surface. Poroid surface fulvous brown when dry; margin buff-yellow, up to 2 mm wide; pores circular to angular, 2–4 per mm, dissepiments very thin, strongly lacerate. Context cinnamon buff, soft corky, thin, up to 1 mm thick, homogeneous. Tubes concolorous to pore surface, slightly darker than context, up to 4 mm long.

HYPHAL STRUCTURE — Hyphal system monomitic; all septa without clamp connections; tissue becoming bloody red but otherwise unchanged in KOH.

CONTEXT — Contextual hyphae pale yellowish, thin- to fairly thick-walled with a wide lumen, occasionally branched, frequently septate, more or less flexuous, loosely interwoven, 4–6  $\mu\text{m}$  in diam.

TUBES — Tramal hyphae hyaline to pale yellowish, thin-walled, rarely branched, frequently simple septate, more or less straight, subparallel along the tubes, 2–4  $\mu\text{m}$  in diam. Setae absent; basidia clavate, with four sterigmata and a simple septum at the base, 10–15  $\times$  4.5–5.5  $\mu\text{m}$ ; basidioles in shape similar to basidia, slightly smaller.

SPORES — Basidiospores oblong ellipsoid, slightly curved, yellowish, fairly thick-walled, smooth, IKI–, moderately CB+, (3.9–)4–4.8(–4.9)  $\times$  (1.9–)2–2.5(–2.6)  $\mu\text{m}$ , L = 4.33 W = 2.15  $\mu\text{m}$ , Q = 2.01 (n = 30/1).

REMARKS — *Phylloporia oblongospora* is characterized by an annual growth, homogenous context, large pores, and oblong ellipsoid basidiospores. It has homogenous context, which is exceptional in *Phylloporia*; however, its hyphal structure, basidiospores, and living environment fit the genus well.

*Phylloporia oblongospora* and *P. fruticum* (Berk. & M.A. Curtis) Ryvar den share very similar pore morphology, but the latter species has distinct duplex context and especially broadly ellipsoid to subglobose basidiospores (3–4.5  $\times$  2.5–3  $\mu\text{m}$ , Wagner & Ryvar den 2002).



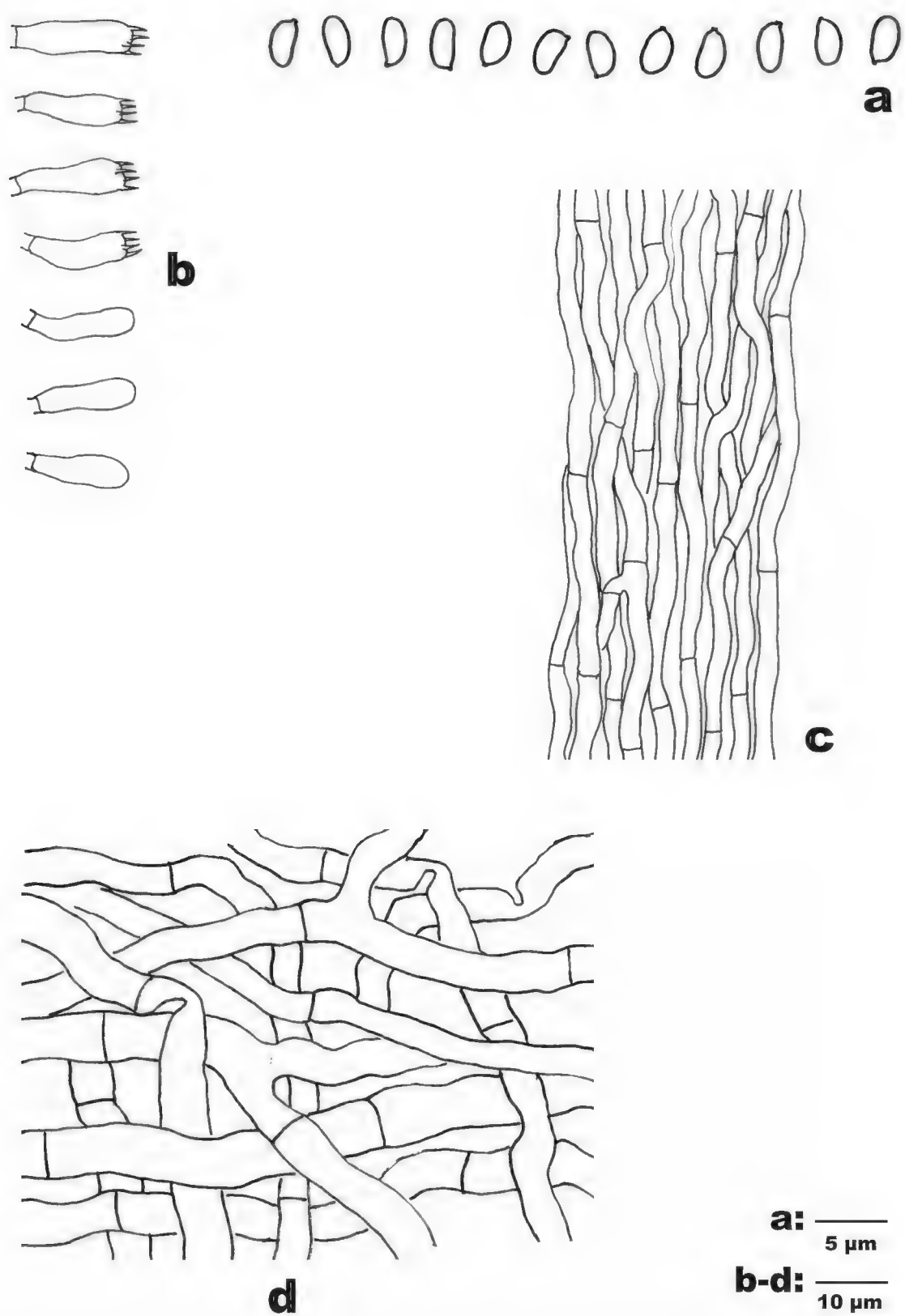


FIG. 2. Microscopic structures of *Phylloporia oblongospora* (drawn from the holotype).  
a: Basidiospores. b: Basidia and basidioles.  
c: Hyphae from tube trama. d: Hyphae from context.

So far, six species in *Phylloporia* have been recorded from China (Dai 1999, Dai et al. 2007a, b, Li et al. 2007, Cui et al. 2008). An identification key to the species of *Phylloporia* occurring in China is provided as following.

Key to species of *Phylloporia* in China

(spore dimensions are provided after species names)

- 1. Basidiocarps perennial, margin acute, hyphae in the upper tomentum  
acyanophilous .....2
- 1. Basidiocarps annual, margin obtuse, hyphae in the tomentum moderately  
cyanophilous .....3
- 2. Pores 6–8 per mm, hyphal system dimitic, cystidioles present  
..... *P. ribis* (Schumach.) Ryvarden  
(2.8–)3–3.8(–4.1) × (1.9–)2–2.6(–2.7) μm,  
L = 3.51 μm, W = 2.22 μm, Q = 1.34–1.58 (n = 60/2)
- 2. Pores 8–11 per mm, hyphal system monomitic, cystidioles absent  
..... *P. pectinata* (Klotzsch) Ryvarden  
(2.4–)2.7–3.3(–3.6) × (1.9–)2–2.5(–2.8) μm,  
L = 2.96 μm, W = 2.21 μm, Q = 1.33–1.35 (n = 60/2)
- 3. Pores 2–4 per mm, context homogeneous; basidiospores oblong ellipsoid  
..... *P. oblongospora*  
(3.9–)4–4.8(–4.9) × (1.9–)2–2.5(–2.6) μm,  
L = 4.33, W = 2.15 μm, Q = 2.01 (n = 30/1).
- 3. Pores 4–9 per mm, context duplex, basidiospores ellipsoid .....4
- 4. Pores 4–6 per mm, basidiospores > 4.6 μm in length .....*P. hainaniana*  
(4.2–)4.6–5.6(–6) × (2.8–)3–3.6(–3.9) μm,  
L = 5 μm, W = 3.11 μm, Q = 1.61 (n = 30/1)
- 4. Pores 6–9 per mm, basidiospores < 4.6 μm in length .....5
- 5. The tomentum up to 1.5 cm thick, concentrically zonate, hyphae of tomentum  
5–9 μm in diam, tramal hyphae distinctly thick-walled, infrequently septate  
..... *P. weberiana* (Bres. & Henn. ex Sacc.) Ryvarden  
(3–)3.4–4.1(–4.5) × (2–)2.2–3(–3.2) μm,  
L = 3.76 μm, W = 2.52 μm, Q = 1.49 (n = 30/1)
- 5. The tomentum up to 0.5 cm thick, azonate, hyphae of tomentum 4–6 μm  
in diam, tramal hyphae fairly thick-walled, frequently septate  
.....*P. bibulosa* (Lloyd) Ryvarden  
3.1–)3.5–4.6(–5) × (2.1–)2.3–3.2(–3.7) μm,  
L = 4.12 μm, W = 2.74 μm, Q = 1.44–1.57 (n = 60/2)

Acknowledgements

We express our gratitude to Prof. Kevin D. Hyde (Mae Fah Luang University, Thailand) for revising the English of the text, and to Drs. Michal Tomšovský (Mendel University, Czech Republic) and Zheng Wang (Yale University, USA) who reviewed the manuscript. The research was financed by the Fundamental Research Funds for the Central Universities (Project No. BLYX200912), the National Natural Science Foundation of China (Project No. 30870013) and the Ministry Science and Technology of China (Project No. 2008BADB0B03).

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## MYCOTAXON

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**Revised description of *Pseudocercospora cornicola***

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**Abstract** — *Pseudocercospora cornicola* (basionym *Cercospora cornicola*), the causal organism of cercospora leaf spot on flowering dogwood, was redescribed and illustrated from its type material and from fresh collections made in Alabama, where it is of common occurrence. Sequence data was obtained from four isolates to support the morphological data for species and generic concepts. The appropriateness of its generic classification is discussed and comments are made on previous descriptions and circumscription of the genera *Cercospora*, *Pseudocercospora*, and *Pseudocercosporella*.

**Key words** — *Cornus florida*, hyphomycete, sequence analysis, taxonomy

**Introduction**

The hyphomycetous anamorph *Cercospora cornicola* was first named and described in 1896, based on a collection made the previous year on languishing leaves of *Cornus florida* at Ocean Springs, Mississippi by S.M. Tracy & F.S. Earle. The fungus was described by Tracy & Earle (1896) as follows:

“Epiphyllous, on irregular brown deadened spots without a definite border, 5–10 mm. Hyphae densely clustered from a nodular base, very short, continuous, somewhat flexuous, olivaceous, 11–15 by 3–4  $\mu$ m; conidia slender, thread-like, somewhat curved, mostly continuous, hyaline or light olivaceous, 60–70 by 2–3  $\mu$ m.”

Type specimens were deposited in the herbaria of Cornell University (CUP), the U.S. Department of Agriculture (BPI), Rutgers College (RUT), Columbia University, and Harvard University (FH).

Chupp (1953) emended Tracy & Earle’s description of *C. cornicola*, describing it thusly:

“Leaf spots irregular brown areas without definite borders, 5–10 mm in extent; fruiting epiphyllous; stroma small, dark, globular, 20–40  $\mu$ m in diameter; fascicles dense to very dense; conidiophores very pale



olivaceous brown, delicate, wavy, uniform in width and color, septa not visible, not or rarely mildly geniculate, not branched, rounded tip, spore scars not visible,  $2\text{--}3.5 \times 10\text{--}25 \mu\text{m}$ ; conidia narrowly obclavate, subhyaline to very pale olivaceous, mildly curved, obconic base, sub-acute tip, septa indistinct,  $2\text{--}3 \times 20\text{--}70 \mu\text{m}$ ."

Hosts were listed as *Cornus florida*, *C. officinalis* Siebold & Zucc., *C. controversa* Hemsl., and *Cornus* spp. (Chupp 1953).

The original description of *Cercospora* by Fresenius allowed a broad concept of the genus to be adopted, as a result of which hundreds of species were classified within it. However, it has subsequently been broken down into smaller, more narrowly defined, segregate genera, including *Pseudocercospora* Speg. and *Pseudocercosporella* Deighton. Modern descriptions of these two genera can be found in Ellis (1971) and Braun (1995), respectively. *Cercospora cornicola* lacks the prominent, thickened conidiophore and conidial scars typical of *Cercospora* and hence was reclassified by Guo & Liu (1989) as *Pseudocercospora cornicola*, but it was not given a comprehensive, updated description.

*Pseudocercospora* and *Pseudocercosporella* are closely related genera that are part of a continuum (Braun 1995). The main difference between the two is that *Pseudocercosporella* consists of fungi with colorless conidiophores and conidia and well-developed, hyaline or subhyaline, rarely pigmented, stromata, whereas *Pseudocercospora* species have pigmented conidiophores and conidia (Braun 1995).

*Pseudocercospora cornicola* occurs commonly on living leaves of flowering dogwood (*C. florida*) in the southeastern United States. Records exist of its occurrence in Japan (Chupp 1953) and China (Guo & Liu 1989). Recent fresh collections obtained in Alabama and examination of the type material of *C. cornicola* has allowed the opportunity to provide a more thorough taxonomic description with illustration.

### Taxonomic description

***Pseudocercospora cornicola*** (Tracy & Earle) Y.L. Guo & X.J. Liu,

Mycosystema 2: 232, 1989.

FIG. 1

= *Cercospora cornicola* Tracy & Earle, Bulletin of the Torrey Botanical Club 23:205, 1896.

Leaf spots necrotic lesions, vein-limited, angular, irregularly shaped, up to 10 mm in diameter, and often confluent. Mycelium internal; composed of branched, septate, pale brown hyphae; and  $2\text{--}3 \mu\text{m}$  in diameter. Caespituli epiphyllous, consisting of punctiform fascicles, olivaceous brown, discrete, usually abundant, and gregarious to somewhat scattered. Stroma well-developed; erumpent; partly superficial, partly immersed; pale to mid-brown; composed of densely packed, predominately isodiametric, subglobose to somewhat angular cells; pseudoparenchymatous; and up to  $70 \mu\text{m}$  in diameter. Conidiophores

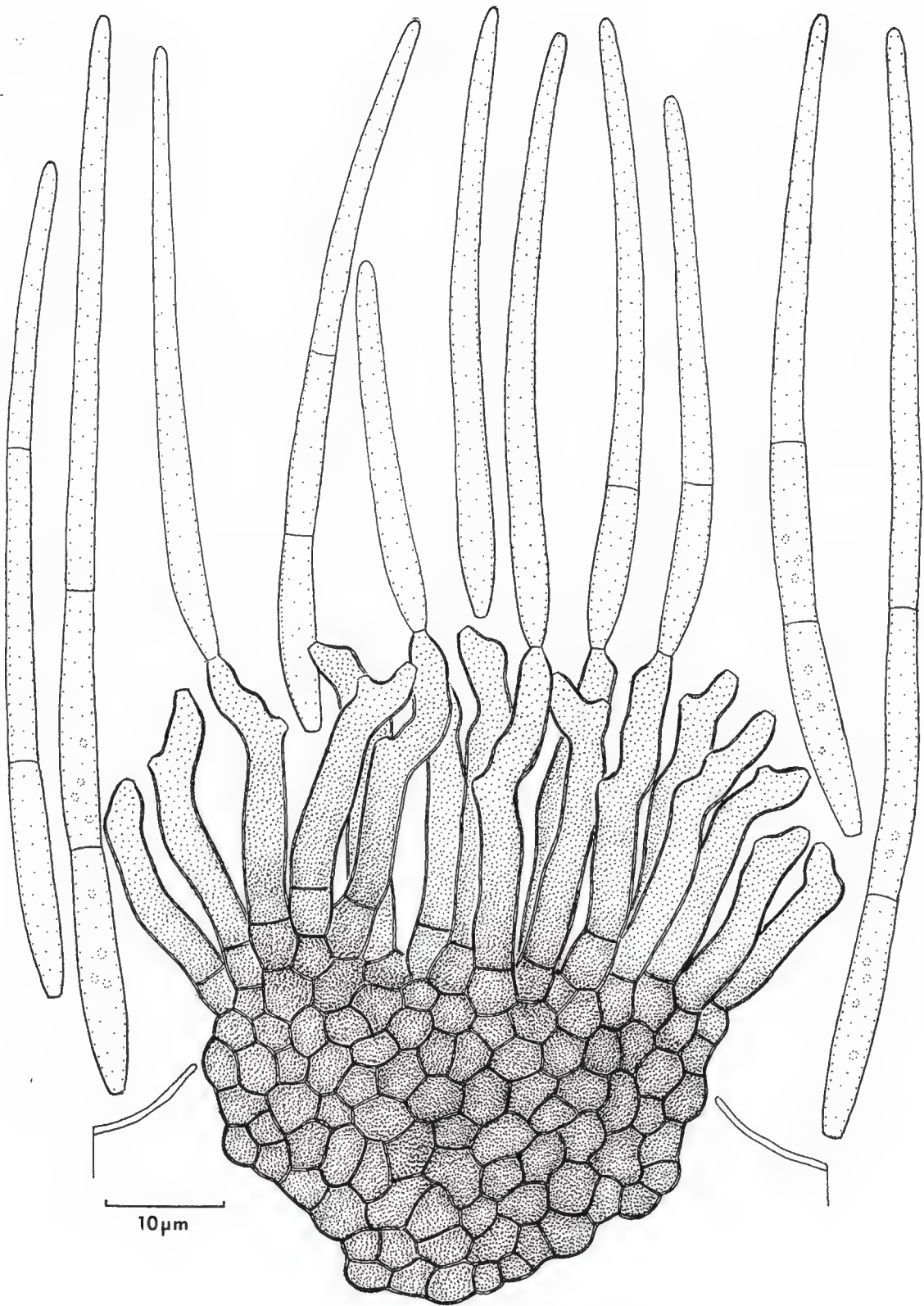


FIGURE 1. *Pseudocercospora cornicola* caespituli.

numerous in dense fascicles, pale olivaceous brown, smooth walled, cylindrical, straight, or slightly curved, becoming geniculate distally with age, usually one septum, 2–3  $\mu\text{m}$  in width, up to 4  $\mu\text{m}$  at the base, and up to 25  $\mu\text{m}$  in length. Conidia narrowly obclavate, hyaline to very pale olivaceous-brown, straight to slightly curved, faintly septate, usually 1–2 septa, sometimes 3 septa, obtuse at apex, truncate at base, 2–3  $\mu\text{m}$  in width, and 20–70  $\mu\text{m}$  in length.

Cosmopolitan on living leaves of *C. florida* L.

COLLECTIONS EXAMINED (all on *Cornus florida*): ALABAMA—Elmore County, Wetumpka: August 31, 2005, K. N. Conner, AUA; Lee County, Auburn: August 31, 2005, K. N. Conner, AUA. MISSISSIPPI—Ocean Springs: September 29, 1895 [CUP-039517, isotype].

### Sequence analysis

Four isolates of *P. cornicola* (collected from Auburn, Lee County and Wetumpka, Elmore County, Alabama) were grown on Acidic Potato Dextrose Agar (APDA) at 30°C for 4 weeks. DNA was extracted with an UltraClean™ Microbial DNA Isolation Kit (MoBio Laboratories, Carlsbad, CA) following manufacturers recommendations. The DNA was amplified using universal fungal primers 2234C and 3126T, designed to amplify the 3' end of the 18S ribosomal RNA gene, internal transcribed spacer 1, 5.8S ribosomal RNA gene, internal transcribed spacer 2, and the 5' end of the 28S ribosomal RNA gene (Ranjard et al. 2001). The PCR products (approximately 500 bp) were sequenced and deposited in GenBank (accessions GU573789, GU573790, GU991657, and GU991658). The four sequences were compared to those present in GenBank using the nucleotide basic local alignment search tool (nBLAST) to support the morphological data and generic placement (Altschul et al. 1990).

### Discussion

Although currently classified in *Pseudocercospora*, *P. cornicola* has some characteristics in common with taxa placed in *Pseudocercospora*, particularly the presence of conidiophores bearing inconspicuous, unthickened, colorless conidial scars and filiform, thin-walled conidia whose base is also unthickened (Braun 1995). On account of this, *Pseudocercospora* might be a more appropriate generic home for this species. However, the stromata and conidiophores are somewhat pigmented and therefore its placement in *Pseudocercospora* is probably warranted. This taxon is, essentially, an entity with features that are intermediate between the two genera. The ITS sequence data showed a 98% similarity between the four isolates and 97% or higher similarity with other *Pseudocercospora* sequences found in GenBank. Furthermore, the *P. cornicola* sequences showed 91% or lower similarity to *Cercospora* sequences and 83%

or lower similarity to *Pseudocercospora* sequences found in GenBank, which supports the generic placement.

The revised description differs notably from previous accounts in that the stroma is well developed and up to 70 µm in diameter, the conidiophores become geniculate distally with age and usually contain one septum, and the conidia are faintly septate, usually containing 1–2 and sometimes 3 septa. With this new description there should be no confusion as to the identity of *P. cornicola* on flowering dogwoods.

### Acknowledgments

We thank Kathie T. Hodge, Cornell University Plant Pathological Herbarium (CUP), for affording us the opportunity to examine the *P. cornicola* isotype. Dr. John M. McKemy, United States Department of Agriculture, and Dr. Richard Baird, Mississippi State University, provided presubmission reviews of the manuscript for which we are grateful.

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## Taxonomic implications of antheridial variability in forty-five watermold isolates: a statistical analysis

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**Abstract** – Morphological variability of sexual features used in watermold identification renders species identifications difficult and calls into question the taxonomic utility of these characters. Herein I have employed chi square statistical analysis to quantify antheridial character state distribution differences between replicate pairs of 38 isolates representing the saprolegniaceous genera *Achlya*, *Saprolegnia*, *Thraustotheca*, and 7 non-sporulating watermolds. Thirty-nine of 45 pairs differed at or below the  $P=.05$  significance level, suggesting that current morphological species concepts are inadequate, at least for *Achlya* and *Saprolegnia*.

**Key words** – *Oomycota*, *Saprolegniaceae*, morphology, systematics

### Introduction

Identification of watermolds (*Saprolegniales*, *Oomycota*) belonging to the genera *Achlya* Nees and *Saprolegnia* Nees has long been problematic owing principally to extensive morphological overlap among recognized species (Johnson et al. 2002). Hulvey et al. (2007) studied this problem in 55 isolates of *Saprolegnia* and demonstrated that little correlation exists between species boundaries based on sexual morphology and those based on gene sequence analysis.

More recently, Sheffer & Padgett (2008) demonstrated that variations in oospore diameter, oospore centricity, antheridial origin, and antheridial appression among subcultures of a single *Saprolegnia* isolate were as great as those that have been used to separate different species in other studies. Their study called into question the taxonomic validity of these sexual characters. The principal unanswered question arising from their report was whether or not the isolate in question was aberrant or demonstrated a degree of variability that applied to the genus or family as a whole. The present study was designed to answer this larger question by assessing the extent of antheridial variability between replicate colonies of 45 isolates belonging to *Achlya* (26 isolates),

*Saprolegnia* (10 isolates), *Thraustotheca* Humphrey (2 isolates), and non-sporulating watermolds (7 isolates).

### Materials and methods

I extracted the data for the current investigation from a much broader study (hereafter referred to as the 'master study') aimed at reevaluating the taxonomic foundation of the family *Saprolegniaceae*. In the course of the master study, approximately 490 watermolds either were acquired from culture collections (the Centraalbureau voor Schimmelcultures [CBS] and the American Type Culture Collection [ATCC]) or isolated from soil samples collected in Italy, Australia, Costa Rica, Canada, Hawaii, and the continental United States. All culture numbers cited herein (TABLE 1) refer to stocks maintained, during the master study, in the watermold culture collection at the University of North Carolina Wilmington.

All isolations from soil were made using standard methods (Johnson 1956, Seymour 1970) as modified below. Soil samples (10 g) were dispensed into disposable 15 × 100 mm Petri dishes, flooded with distilled water (DW), and baited with sterile, shelled hemp seeds (hs). Culture plates then were incubated at room temperature until watermold colonies developed. Axenic cultures subsequently were derived by single spore or hyphal tip isolations from gross cultures and maintained on hs in water and on Difco corn meal agar (CMA) in preparation for microscopic analysis.

Morphological characterization of all axenic isolates required 10 replicate, DW-grown subcultures for each watermold. These were initiated first by infesting 10 sterile hs for 24 h at the edge of CMA-grown colonies then transferring individual, infested hs to separate Petri dishes containing 20 mL of DW. After incubation at room temperature for 24 to 48 h isolates were identified to genus by observing zoosporangial discharge from 10 primary sporangia. Incubation then continued for up to 14 days until mature oogonia and antheridia were visible.

As asexual and sexual features matured through time, individual colonies (of the 10 replicates for each isolate) were harvested for morphological characterization; qualitative data were recorded on separate data sheets (one data sheet per replicate subculture). These observations were made using Olympus phase contrast microscopes with 400× magnification. During data collection we attempted to record 50 observations for all sexual characters presented by a particular colony at the time of harvest, but rarely were unobstructed views of this number available.

Of the 490 watermolds acquired in the master study we identified all that produced zoosporangia to genus. About half of the axenic cultures subsequently produced sexual features, but few were good fits to described species. I limited isolates for the present statistical analysis to those with 50 character state observations of the same sexual character on each of two separate data sheets (i.e. from two separate replicate colonies of the same isolate). Ultimately only 45 isolates met this criterion and of those only two sexual characters (antheridial origin and antheridial appression) consistently presented the required sample sizes. Thirty-five cultures qualified for statistical analysis for both antheridial characters and the remaining 10 qualified for one character. Hereafter the two replicates for each qualifying culture are referred to as a 'replicate pair' (RP).

All RPs in the test pool presented 3 character states for antheridial origin – monoclinal, diclinal, and androgynous – , and 3 for antheridial appression – apical, lateral, and projections (illustrations in Johnson 1956). Members of each RP were compared for uniformity of character state distribution (for both characters) using Chi square statistical analysis. For calculation purposes I used the mean value (per character state) as the ‘expected’ value for the particular RP. This necessitated doubling each ‘calculated’ Chi square to derive the value used for comparison to the appropriate tabular Chi square value. I considered  $P=.05$  to be the maximum level for statistical significance.

## Results

TABLE 1 presents results of Chi square comparisons of all 45 RPs. In all cases I made the conservative assumption that any RP for which one character had insufficient data for comparison (less than 50 observations) did not differ for that character. This being the case, when results for all genera were combined I found that only 6 of 45 RPs had no significant differences for either character. Of the remaining 39 all exhibited differences in at least one character. Furthermore 15 of 45 exhibited significant differences for both characters.

Separating results by genus revealed that all 10 RPs of *Saprolegnia* differed with respect to one or both characters, both RPs of *Thraustotheca* differed for one character, and all seven non-sporulating RPs differed for one or both characters. *Achlya* RPs were the least variable, yet 12 of 26 pairs differed with respect to one character while 8 differed for both.

## Discussion

In light of the present data (TABLE 1), it is apparent that the statistically significant variability reported by Sheffer & Padgett (2008) was not aberrant but may be typical not only for *Saprolegnia* but also for *Achlya*. I am keenly aware that drawing sweeping conclusions based on data for only two sexual characters is risky. Consequently, I visually inspected raw data (from the master study described above) for other watermolds that did not qualify for the present analysis and found comparable variability in oogonial and oospore characters.

I carefully reviewed historical monographs of saprolegniaceous genera (Coker 1923, Coker & Matthews 1937, Johnson 1956, Scott 1961, Seymour 1970) and found no mention of statistical tests ever having been applied to assess variability of taxonomic characters. Clearly results reported herein demonstrate that this omission represents a serious taxonomic problem that introduces an unacceptable level of subjectivity into identifications of *Achlya* and *Saprolegnia* isolates.

New watermold species currently are being erected at an alarmingly rapid pace (e.g. Steciow 2001a,b, 2002, 2003a,b, Steciow & Elides 2002a,b,c, Steciow



TABLE 1. Chi square significance levels per replicate pair for antheridial characters

GENUS	STOCK CULTURE NUMBER <sup>^</sup>	ANTHERIDIAL ORIGIN	ANTHERIDIAL APPRESSION
<i>Achlya</i>	223	**	**
<i>Achlya</i>	234	ID <sup>b</sup>	**
<i>Achlya</i>	243	ID	***
<i>Achlya</i>	246	P>.25	***
<i>Achlya</i>	247	P>.1	***
<i>Achlya</i>	267	*	***
<i>Achlya</i>	276	**	***
<i>Achlya</i>	281	***	P>.25
<i>Achlya</i>	287	P>.25	***
<i>Achlya</i>	313	ID	***
<i>Achlya</i>	326	*	***
<i>Achlya</i>	342	P>.25	***
<i>Achlya</i>	347	*	***
<i>Achlya</i>	362	***	***
<i>Achlya</i>	367	*	ID
<i>Achlya</i>	418	**	***
<i>Achlya</i>	451	**	P>.1
<i>Achlya</i>	455	**	P>.1
<i>Achlya</i>	456	P>.1	P>.25
<i>Achlya</i>	460	P>.1	P>.1
<i>Achlya</i>	462	P>.1	P>.25
<i>Achlya</i>	463	P>.05	P>.25
<i>Achlya</i>	465	P>.25	***
<i>Achlya</i>	469	P>.25	ID
<i>Achlya</i>	485	P>.25	P>.1
<i>Achlya</i>	487	**	*
<i>Saprolegnia</i>	105	ID	***
<i>Saprolegnia</i>	217	***	ID
<i>Saprolegnia</i>	254	***	P>.25
<i>Saprolegnia</i>	257	P>.25	*
<i>Saprolegnia</i>	262	**	***
<i>Saprolegnia</i>	280	**	P>.25
<i>Saprolegnia</i>	284	***	P>.25
<i>Saprolegnia</i>	361	***	***
<i>Saprolegnia</i>	383	*	P>.1
<i>Saprolegnia</i>	472	**	***
<i>Thraustotheca</i>	60	ID	***
<i>Thraustotheca</i>	325	ID	***
Unknown <sup>a</sup>	277	P>.25	*
Unknown	291	***	ID
Unknown	292	**	***
Unknown	299	***	**
Unknown	380	*	*
Unknown	382	P>.1	***
Unknown	424	***	*

<sup>^</sup> UNC-W watermold culture collection. <sup>a</sup> Unknowns did not produce zoosporangia. <sup>b</sup> ID = insufficient data for statistical comparison, \* indicates P≤.05, \*\* indicates P≤.01, \*\*\* indicates P≤.001.

et al. 2007, Steciow & Marano, 2008, Paul & Steciow 2004, 2008, Johnson et al. 2005, Amal et al. 2006, Sati & Paliwal 2006), yet no descriptions have been accompanied by morphological variability assessments. Continuing this practice inevitably will render watermold taxonomy progressively more problematic.

Recent literature (e.g. Leclerc et al. 2000, Bouzenzana et al. 2006, Hulvey et al. 2007, Dieguez-Uribeondo et al. 2007, Fregeneda-Grandes et al. 2007) reflects a gratifying expansion both of biochemical and gene sequence information that no doubt will be of great value in comprehensive revision of *Oomycete* taxonomy. Such studies, however, represent only the start of a necessary baseline that must develop more fully before meaningful revision can emerge. Few scientists would argue with the paradigm that genes determine biochemistry, which determines morphology. I must infer, therefore, that the variability reported herein reflects some currently unknown disconnect between genes and morphology that renders present concepts of *Achlya* and *Saprolegnia* species inadequate.

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## MYCOTAXON

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**New records of lichens and lichenicolous fungi from Iran  
and their biogeographical significance**

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**Abstract** — In this paper, 80 lichen taxa and 9 lichenicolous fungi are reported as new to Iran. These include a tropical element represented by *Lithothelium obtectum* and *Melanotopelia africana*, and a North American element with *Lecanora flowersiana*, *L. juniperina*, *L. percrenata*, and *L. wetmorei*. The full checklist is available on <http://www.mycotaxon.com>

**Key words** — lichenized fungi, new species records, biogeography

**Introduction**

The recently revised checklist of lichenized, lichenicolous, and allied fungi for Iran (Seaward et al. 2008) includes 632 records based on literature records and voucher material, which means an increase of 224 species as compared to an earlier list (Seaward et al. 2004). Nevertheless, the exploration of the Iranian lichen flora appears far from being complete, with Valadbeigi et al. (2010) and Haji Moniri & Sipman (2009) having added another 24 species new to this region. The present paper reports 89 additional lichens and lichenicolous fungi new to Iran.

Iran is one of the world's most mountainous countries and largely occupied by the Iranian Plateau. Extended lowlands exist only along the coasts of the Caspian Sea and in Khuzestan. The specimens for the present study were collected from six different provinces (FIG. 1) in areas with a wide range of ecological characteristics.





Fig. 1. Study areas in the six provinces:  
I, Azarbaijan; II, Gilan; III, Gorgan; IV, Hamedan; V, Ilam; VI–VIII, Mazandaran.

### Materials and methods

The study is based on material collected by the first author during 2004–2009. The specimens are deposited in TARI (the Research Institute of Forests and Rangelands, Tehran), with some duplicates in B (Botanischer Garten und Botanisches Museum Berlin) and the private herbarium of the first author (VH). The morphology of all specimens was studied with a stereomicroscope. The chemistry was mostly investigated by using standard spot tests. Identifications were confirmed by comparison with specimens kept in the herbarium of B or by consultation with specialists. In critical cases, chemical analyses were carried out using TLC following Orange et al. (2001), using Merck silica gel 60 F254 pre-coated glass TLC plates in solvent systems A, B', and C. Identification of the substances was confirmed by running the extract next to a reference sample with known chemistry (co-chromatography).

### Phytogeographical discussion

These reports confirm that the lichen flora of Iran is mostly composed of boreal, mediterranean, and central-asian phytogeographical elements (often

widespread, as would be expected) but that it includes other lichen-floristic elements as well.

Species with a major distribution in the Himalaya and East Asia have been previously reported, such as *Cladonia awasthiana* Ahti & Upreti (Seaward et al. 2004, Ahti & Sohrabi 2006) and *Leptogium trichophorum* Müll. Arg. (Haji Moniri & Sipman 2009). Both occur in the northern mountain range, which can be considered a continuation of the Himalayas. *Cladonia awasthiana* seems widespread in the Hyrcanian forest area, while *L. trichophorum* is known so far only from a single collection around the 2500 m elevation in Northern Khorasan.

*Pyrgidium montelicum* (Beltr.) Tibell, reported by Seaward et al. (2004), was the first indication of a tropical element in the Iranian lichen flora. This species is mainly known from the Palaeotropics, although with outliers reported as far north as Italy (Tibell 1982, 1996). Three additional species with a predominantly tropical distribution are reported here from Iran: *Lithothelium obtectum* (Müll. Arg.) Aptroot, hitherto known to be pantropical and very common in India (Aptroot 1991); *Melanotopelia africana* Sérus. et al., known previously from tropical continental Africa, La Réunion (Sérusiaux et al. 2009) and Borneo (Sipman 31228 in herb. B); and *Siphula decumbens* Nyl., known from the Neotropics and the Palaeotropics with an extension to Japan (Kantvilas et al. 2005). All were found in Iran along the Caspian coast in the Hyrcanian forest zone. However, the altitudinal range is 450–2600 m, and not all grow in forest habitats.

Some additional species appear to represent a North American element. This element had earlier been indicated by a group of lichens that are rather common in Iran and surroundings, although they are absent from Western Europe: *Ramalina sinensis* Jatta, *Lecanora thysanophora* R.C. Harris, and *Pyrenula subelliptica* (Tuck.) R.C. Harris (Seaward et al. 2008; for extra-Iranian distribution see Purvis et al. 1992, Brodo et al. 2001, Harris 1989). Based on the treatment of the North American representatives of the *Lecanora dispersa* group by Śliwa (2007), several further species are reported here: *Lecanora flowersiana* H. Magn., *L. juniperina* Śliwa, *L. percrenata* H. Magn., and *L. wetmorei* Śliwa. Of these only *L. percrenata* had been previously reported from outside North America, from Central Asia (Śliwa 2007). The species of this element seem to be widely distributed in Iran.

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## MYCOTAXON

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**First records of *Clathrus* (Phallaceae, Agaricomycetes)  
from the Northeast Region of Brazil**E.P. FAZOLINO<sup>1</sup>, L. TRIERVEILER-PEREIRA<sup>2</sup>, F.D. CALONGE<sup>3</sup> & I.G. BASEIA<sup>4</sup><sup>1</sup>*edufazol@yahoo.com.br* & <sup>4</sup>*baseia@pesquisador.cnpq.br**Depto. Botânica, Ecologia e Zoologia, Universidade Federal do Rio Grande do Norte  
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**Abstract** — Two *Clathrus* specimens were collected in Northeastern Brazil during the rainy season in 2008. One specimen was identified as *C. chrysomycelinus* and the other is described as a new species, *C. cristatus*, which is distinguished by its pale red to pink receptacle and crests along the edge. Full descriptions with illustrations of the collected specimens and a key to species of *Clathrus* from Brazil are provided.

**Key words** — *Clathraceae*, gasteromycetes, Neotropical mycodiversity

**Introduction**

*Clathrus* P. Micheli ex L. was validated in 1753; the type species is *C. ruber* P. Micheli ex Pers. According to the most recent edition of the Dictionary of Fungi, this genus embraces 16 species, which are widespread in tropical and subtropical areas (Kirk et al. 2008).

In his treatment of the *Clathraceae*, Dring (1980) recognized 15 species and a specimen that he labelled “*Clathrus* species 1” due to the fact of having studied only a single basidioma preserved in spirit on which to base his formal diagnosis. García & López (1995) later proposed *C. mexicanus* as a new species; examination of the type material by one of us (Dr. Calonge), however, led to the conclusion that its taxonomic status is doubtful, because the basidioma

**NOTE** — MYCOTAXON prepared this PDF with color plates for the author. The original print version was published with halftone (grayscale) plates.



is abnormal and does not give enough information to clarify its real identity (Calonge et al. 2004). Other recent additions to the genus are *C. xiningensis* (H.A. Wen) B. Liu. (Fan et al. 1994) and *C. hainanensis* X.L. Wu (Wu 1998).

The genus is characterized by a latticed, clathrate receptacle composed of hollow, tubular arms that arise from the basal tissue within the volva (Miller & Miller 1988). The deliquescent gleba usually develops on the inner side of the receptacle and the basidiospores are elliptical and smooth (Dring 1980). The unpleasant odor produced by the gleba attracts flies and other insects, contributing to basidiospore dissemination (Maldonado-Ramírez & Torres-Pratts 2005).

The existence of the genus in Brazil has been known since the 19th century. Fidalgo (1974) reported that a *Clathrus* specimen gathered in 1826 by William John Burchell comprised the first collection of a gasteroid fungus in the country. To date, five *Clathrus* species have been recorded from Brazil (see key below) and their range is so far restricted to the Southern regions (Trierveiler-Pereira & Baseia 2009). *Clathrus chrysomycelinus*, which is the species with the widest distribution in the country, has been recorded from four states: Rio Grande do Sul (Rick 1961), Santa Catarina (Möller 1895), Paraná (de Meijer 2006), and São Paulo (Bononi et al. 1984). Two other species recorded from Brazil, *C. americanus* Lloyd and *C. pseudocrispus* Lloyd, are considered synonyms of *C. crispus* Turpin (Dring 1980).

During recent field expeditions in preserved areas of Northeastern Brazil, two distinct species of *Clathrus* were collected, one of them new to science. The purpose of this study is to present full descriptions and photos of these species and an identification key to species of *Clathrus* recorded from Brazil.

## Materials and methods

Gasteromycete collection was carried out during the rainy season of 2008 (March–August) in preserved forests areas of Northeastern Brazil. RPPN Fazenda Tamanduá (7°00'35"S, 37°23'50"W) is a 325 ha remnant of 'caatinga' (xeric shrubland and thorn forest), located in the city of Santa Terezinha, state of Paraíba. Parque Ecológico João de Vasconcelos Sobrinho (8°17'00"S, 35°58'3"W), also known as 'Brejo dos Cavalos', is a 359 ha remnant of the Atlantic rain forest located in the city of Caruaru, state of Pernambuco.

Basidiomata were examined and photographed in the field. A taxonomic study was performed by observing macro and microscopic features according to Miller & Miller (1988) and Dring (1980). For scanning electron microscopy (SEM), a few drops of isopropyl alcohol were added to gleba samples, coated with gold-palladium on an Ion Sputter Coater, and observed under a Shimadzu SSX-550 scanning electron microscope. Colours were coded according to Kornerup & Wanscher (1978), with the indication "KW" bracketed in the text, and simultaneously described. Vouchers were dried slowly and are kept in the herbaria UFRN-fungi and URM (Holmgren & Holmgren 1998).





FIGURE 1. *Clathrus cristatus*. Basidiome in situ (scale bar = 2 cm).

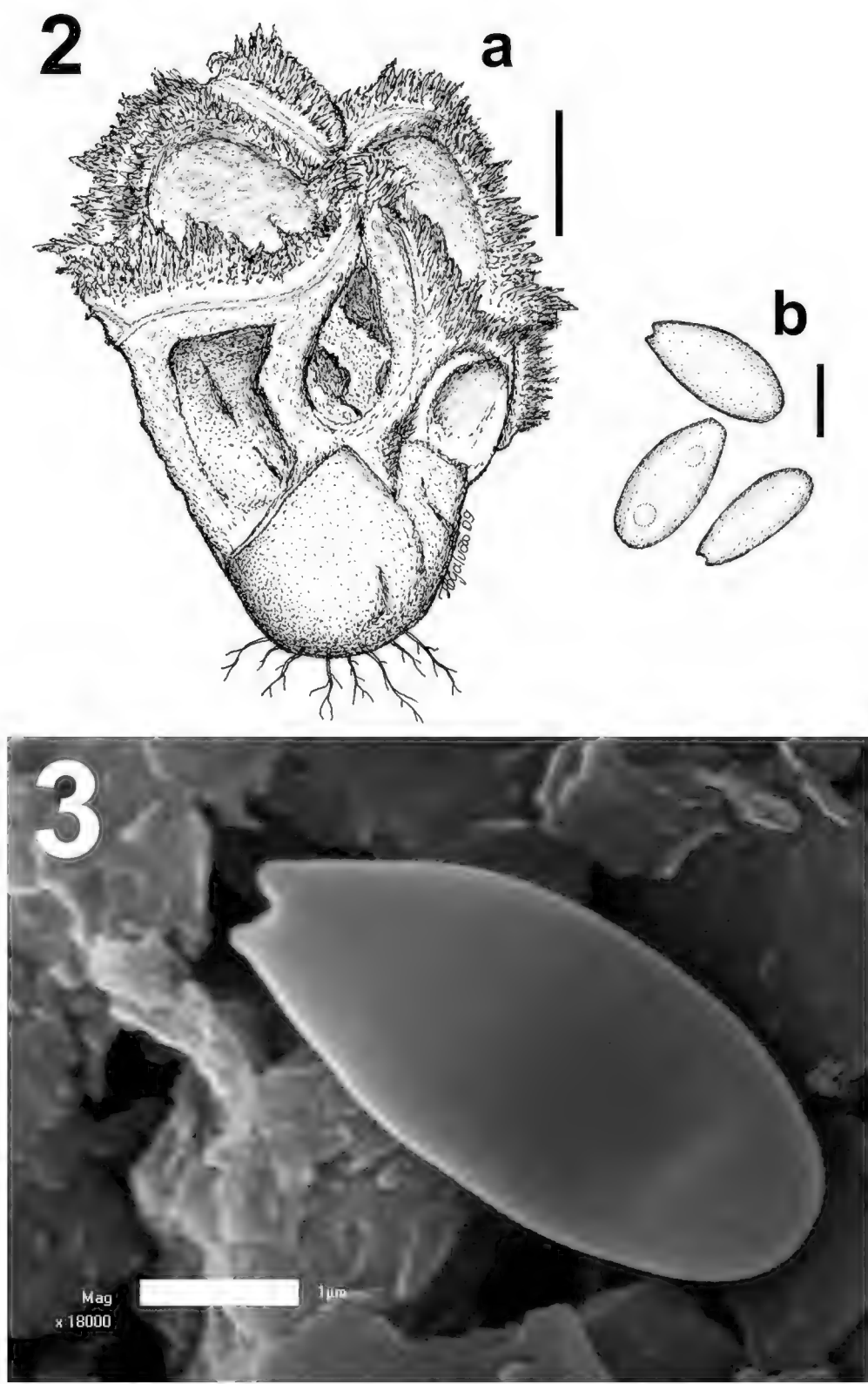
**Taxonomy**

*Clathrus cristatus* Fazolino, Calonge & Baseia, sp. nov.

FIGS 1–3

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*Volva* 1.5–2 cm diam., subglobosa, brunnea-pallida, reticulata, irregulariter dehiscentem, rhizomorphis basalis albis. *Receptaculo* 5 cm alto, 3 cm lato; obovoideo, clathrato cum rami in dispositio symmetricus, roseus in parte externa et scarlatinus ad facies interna, margine cristata. *Gleba* in facies interna ad rami, olivaceus, odore grato; basidiosporis 3.5–5 × 1.5–2 micra, cylindrico-ellipsoideis, chlorohyalinis, laevis.



FIGURES 2–3. *Clathrus cristatus*.

2a. Basidiome (scale bar = 1 cm), 2b. Basidiospores (scale bar = 2 μm).  
3. SEM of basidiospore (scale bar = 1 μm).

TYPE — BRAZIL. PARAÍBA: Santa Terezinha. RPPN Fazenda do Tamanduá. col. E.P. Fazolino 068. 23.III.2008 (*UFRN-FUNGI* 0492, **holotype**).  
ETYMOLOGY — *cristatus* refers to the presence of crests on the edge of the top network.

Volva 1.5–2 cm diam., subglobose, light brown (KW 5D7), with a network of grooves, rooting at the base by several whitish hyphal strands (rhizomorphs); dehiscence by irregular splitting of the apex. Receptacle 5 cm high  $\times$  3 cm diam, obovoid, clathrate with a more or less symmetrical arrangement of the arms to give an irregular network of about eight meshes, with longitudinal grooves at the top ones (FIG. 1, 2), arms 4–6 mm in length, fused at the base, pastel red (KW 10A4) on the outside, shading to red (KW 10A7) within, transverse section of an arm shows two flattened tubes; upper meshes are surrounded by a fringe of crests, crests 1–5 mm long. Gleba borne on the inner face of the arms, distributed all along the arms, olivaceous (KW 3F7), odour of cheese; basidiospores  $3.5\text{--}5 \times 1.5\text{--}2 \mu\text{m}$ , cylindric-ellipsoid, faintly greenish tinted (chlorohyaline), smooth (FIG. 1, 3), at high magnification the surface may appear rugulose, but this is an artifact.

HABITAT — growing solitary on sandy soil.

TAXONOMIC REMARKS — *Clathrus cristatus* is distinguished by its pale red to pink receptacle and crests along the arms edges. *Clathrus preussii* Henn. also shows a fringe of teeth along the edge but these are fewer, smaller and shorter and the receptacle is dirty white. *Clathrus cristatus* arms may also resemble *Laternea pusilla* Berk. & M.A. Curtis, but a careful analysis will show that the receptacle morphology is quite different between the two species.

*Clathrus chrysomycelinus* Möller, Bot. Mitt. Trop. 7: 22 (1895).

FIG 4

MATERIAL EXAMINED — BRAZIL. PERNAMBUCO: Caruaru. Parque Ecológico João de Vasconcelos Sobrinho, col. L. Trierveiler-Pereira & al., 105. 17.VI.2008 (URM 80094).

Volva 1.9 cm high  $\times$  2.9 cm diam, ellipsoid, external layer dark brown (KW 6F4), internal layer yellowish grey (KW 3B2), content gelatinous, rotting at the base by a central rhizomorph, up to 9 cm long, dull yellow (KW 3B4). Receptacle 5.4 cm high  $\times$  4 cm diam, subglobose to obovate, meshes more or less hexagonal, isodiametric in the upper part and elongates below, where the arms are fused and form a short stipe, arms very fragile, slender and flattened (Fig. 4), white (KW 3A1) to pale yellow (KW 4A3), stipe 1.5 cm high  $\times$  1 cm diam. Gleba restrict to glebifers that are situated at the arms junctions, olive (KW 2F6), foetid; basidiospores  $3.5\text{--}4.5 \times 1.5\text{--}2 \mu\text{m}$ , ellipsoid, chlorohyaline, smooth.

HABITAT — growing solitary on soil among litter.

TAXONOMIC REMARKS — Due to its white color, *C. chrysomycelinus* resembles *C. preussii* but lacks the fringe of teeth along the edge. The species may also be confused with the two *Ileodictyon* species: *I. cibarium* Tul. & C. Tul. and *I. gracile* Berk., since the receptacle color and the arm morphology are somewhat





FIGURE 4. *Clathrus chrysomycelinus*. Basidiome in situ (scale bar = 2 cm).

similar. However, in *Ileodictyon* the receptacle arms are not fused to form a short stipe and the whole receptacle occasionally becomes detached from the volva. Moreover, *Ileodictyon* species have a gelatinous receptacle and simple tubular (circular in trans-section) arms without dorsiventral differentiation (Dring 1980, Miller & Miller 1988).

**Key to *Clathrus* species recorded from Brazil**

- 1a. Receptacle white, yellowish white to pale yellow ..... 2
- 1b. Receptacle bright red, reddish orange to very pale red ..... 3
- 2a. Arms with a fringe of small membranous teeth along the edge ..... *C. preussii*
- 2b. Arms without a fringe of membranous teeth ..... *C. chrysomycelinus*

- 3a. Receptacle pale red to pink, dense membranous teeth along the edge . . . *C. cristatus*
- 3b. Receptacle bright red to reddish orange, arms without membranous teeth . . . . . 4
- 4a. Receptacle formed by 2-5 thick, columnar arms, not forming meshes  
    *C. columnatus*
- 4b. Receptacle arms forming meshes . . . . . 5
- 5a. Arms massive, up to 1 cm wide, triangular in transaction, meshes surrounded by  
    gleba, gleba forming a crown . . . . . *C. crispus*
- 5b. Arms slender, more or less circular in transaction, meshes without gleba forming a  
    crown . . . . . *C. pusillus*

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## MYCOTAXON

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***Cladonia*, *Lecanographa*, *Ochrolechia*, and *Placidium*  
species new to Turkey**

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**Abstract**—*Cladonia dahlia*na, *Lecanographa grumulosa*, *Ochrolechia inaequatula* and *Placidium imbecillum* are reported for the first time from Turkey. For each species a short description is presented.

**Key Words**—biodiversity, biota, Giresun, lichen, new record

**Introduction**

Studies on the lichen biota of Turkey are not as extensive as in many European countries. In the last two years, many new lichen species were reported for the lichen biota of Turkey (e.g. Candan & Özdemir Türk 2008, Çobanoğlu et al. 2008, Halici & Aksoy 2009, Kinalioğlu 2009, Öztürk & Güvenç 2010, Yazıcı & Aptroot 2008). So far a total of 518 species have been reported from Trabzon and 431 from Giresun province. The present paper is a further contribution to ongoing lichens exploration in the country.

**Materials and methods**

Specimens were collected in Trabzon and Giresun provinces between year 2005 and 2007. They were identified with various lichen guides (e.g. Brodo et al. 2001, Purvis et al. 1992, Wirth 1995) and determined by H. Sipman. Vouchers are preserved in the herbarium of the Faculty of Science and Arts, Giresun University, Giresun, Turkey; with some duplicates in personal herbarium of H. Sipman. The accession numbers of the collections are given in parentheses after the locality details.

**Species recorded*****Cladonia dahlia*na** Kristinsson

FIG. 1

Primary thallus squamulose dominant, 1–5 mm broad, 3–10 mm long, incised, the surface mostly finely rugose, greenish above, white below. Podetia

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FIG. 1. *Cladonia dahliaana*, habitus. Scale: 1 mm.

up to 4–5 mm tall, corticated, green, gradually tapered towards base. Cups to 4 mm wide, generally dentate at the rim. Apothecia brown, on the cup margins. Medulla K+ yellow, PD+ yellow.

SPECIMEN EXAMINED: Giresun, Dereli, Karagöl mountains, 40°35'51"N, 38°10'30"E, 3050 m, 29 Jul. 2007, on soil, *det. H. Sipman*, (*Kınalıoğlu* 1575).

Known from Iceland, Greenland, Baffin Island on the steep soil banks and hillsides or in the steep sides of snow patches facing south (Kristinsson 1974). In Turkey the specimen was collected from soil at high altitude.

A detailed description of northern European material is provided by Kristinsson (1974).

DISCUSSION: The Turkish material differs from the northern European specimens by having podetia and a wider primary squamulose thallus.

*Lecanographa grumulosa* (Dufour) Egea & Torrente

FIG. 2

Thallus crustose, greyish, mostly thick, cracked-areolate. Apothecia 0.3–1.5 mm diam, black, sessile when old, roundish to ellipsoid: disc plane, white pruinose, crenulate margins. Asci 8-spored, grumulosa type. Ascospores 13–18 × 3–4 µm in size, colourless, 3–4-celled when young and 5–6-celled when mature. Pycnidia not observed. Thallus and apothecial pruina C+.



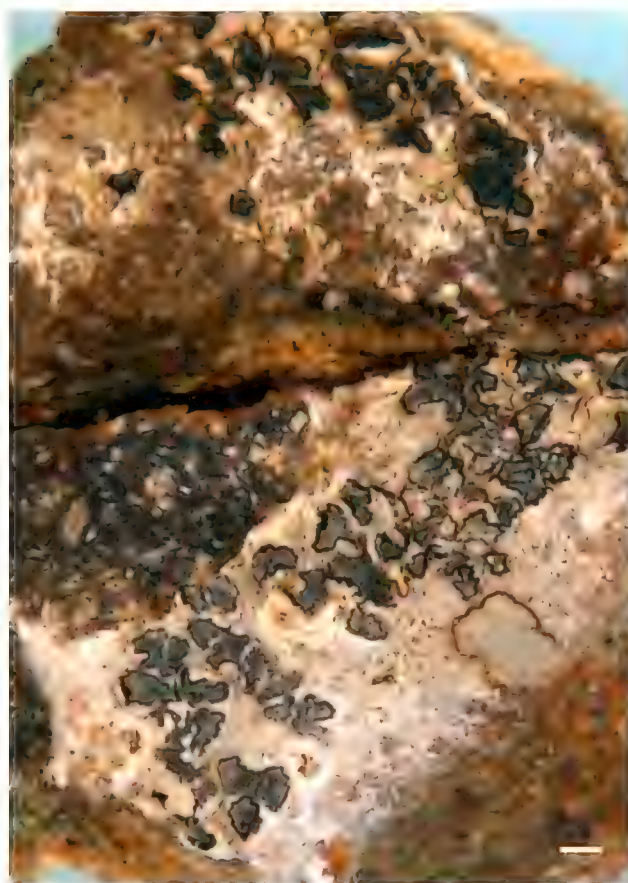


FIG. 2. *Lecanographa grumulosa*, habitus. Scale: 1 mm.

SPECIMEN EXAMINED: Giresun, Gülburnu, sea shore, 40°57'50"N, 38°39'14"E, 1 m, 10 June 2006, on siliceous rock, det. H. Sipman, (Kınalıoğlu 1462). Giresun, Keşap, Değirmenağzı village, 40°58'2"N, 38°38'33"E, 12 m, 12 Dec. 2006, on siliceous rock, det. H. Sipman, (Kınalıoğlu 1511).

Known from Europe on dry  $\pm$  calcareous rocks and mortar, often on sheltered underhangs and shaded walls (Purvis et al. 1992, Egea et al. 1993). In Turkey the specimens were collected only from siliceous rock.

A detailed description of European material (as *Lecaxanactis grumulosa*) is provided by Purvis et al. (1992) and Egea et al. (1993).

DISCUSSION: The Turkish representatives of *Lecanographa grumulosa* differ from European specimens by larger apothecia and slightly larger ascospores. (Egea et al. (1993) cite ascospores as  $12-17(-19) \times 3-4 \mu\text{m}$ , although Purvis et al. (1992) list sizes up to  $14-23 \times 3-4(-5) \mu\text{m}$ ). The Turkish collection differs ecologically in occurring only on siliceous rock at coastal localities.

*Ochrolechia inaequatula* (Nyl.) Zahlbr.

FIG. 3

Thallus thick, uniformly grey-white. Soralia to 1.5 mm diam., sorediata coarse. Photobiont chlorococcoid. Apothecia not observed. Thallus PD+ pale orange.



FIG. 3. *Ochrolechia inaequatula*, habitus. Scale: 1 mm.

SPECIMEN EXAMINED: Trabzon Araklı, S of Kızılkaya Yaylası, 40°40' 21"N, 40°01'24"E, 2350 m, 18 Aug. 2005, on moss, det. H. Sipman, (Kınalıoğlu 1474).

Known from Scotland and Scandinavia on the bryophyte mats on exposed mountain ridges and on tops of boulders (Purvis et al. 1992). In Turkey the specimen was collected from moss at high altitude.

A detailed description of British material is provided by Purvis et al. (1992).

DISCUSSION: The soralia are smaller in the Turkish specimen than in the British material. Original descriptions of this species report soralia up to 2–3 mm diam. (Purvis et al. 1992).

### *Placidium imbecillum* (Breuss) Breuss

FIG. 4

Thallus squamulose, squamules 3–5 mm wide, adpressed to the substratum, dark brown or with a reddish tinge. Perithecia frequent, black, half immersed. Ascospores *colourless*, 13–17 × 6–7.5 µm. Thallus C–, K–, KC–, PD–.

SPECIMEN EXAMINED: Trabzon, Araklı, SE of Paskalar Yaylası, 40°40'03"N, 40°01'41"E, 2400 m, 17 Aug. 2005, on soil, det. H. Sipman, (Kınalıoğlu 1471)

Known from western Europe (Austrian Alps) and several isolated stations in southern Europe on soil (Nimis & Martellos 2004, Breuss 1990). In Turkey the specimen was also collected from soil.



A detailed description of Italian material (as *Catapyrenium imbecillum*) is provided by Nimis & Martellos (2004).

**DISCUSSION:** The squamules and ascospores of the Turkish material are slightly smaller than in the Italian collection. In the Italian specimen the squamule sizes are (2)3–6 mm wide, and the ascospores sizes are (12)14–18 × 6–8 µm.



FIG. 4. *Placidium imbecillum*, habitus. Scale: 1 mm.

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## MYCOTAXON

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**New species of *Hypoxylon*  
from western Europe and Ethiopia**JACQUES FOURNIER<sup>1</sup>*jacques.fournier@club-internet.fr*<sup>1</sup>*Las Muros, F-09420, Rimont, France*BÄRBEL KÖPCKE<sup>2</sup> & MARC STADLER<sup>2,3,\*</sup>*baerbel.koepcke@intermed-discovery.com & marc.stadler@t-online.de*<sup>2</sup>*InterMed Discovery GmbH, Otto-Hahn-Straße 15**D-44227 Dortmund, Germany*<sup>3</sup>*University of Bayreuth, Dept. Mycology**Universitätsstraße 30, D-95540 Bayreuth, Germany*

**Abstract** — Three new species of *Hypoxylon* are described from France, Portugal, and the United Kingdom based on new combinations of teleomorphic morphology. *Hypoxylon fuscoide*s is related to *H. fuscum* but differs in having purple pigments. *Hypoxylon lusitanicum* is similar to *H. perforatum* but differs in having orange stromatal pigments. *Hypoxylon gibriacense* features glomerulate stromata and resembles the American *H. shearii* but has discoid ostiolar areas and different ascospores. In this context, *H. addis*, collected from Ethiopia, is also newly described because it appears morphologically similar to *H. gibriacense*. Their secondary metabolite profiles, as inferred from high performance liquid chromatography coupled with diode array detection and mass spectrometric detection (HPLC-DAD/MS), confirm their uniqueness as compared to related species. Lecanoric acid (widely distributed in lichenized ascomycetes) is revealed to as the major stromatal metabolite of *H. addis* and is for the first time reported as present in a xylariaceous species. A new key to European *Hypoxylon* species is provided.

**Key words** — *Xylariaceae*, chemotaxonomy, systematics, pyrenomycetes

**Introduction**

*Hypoxylon* Bull. has traditionally comprised the largest genus of family *Xylariaceae* with (fide Index Fungorum) over 1100 epithets associated with the generic name. The revision by Ju & Rogers (1996) introduced new species concepts based on a combination of teleomorphic and anamorphic characters

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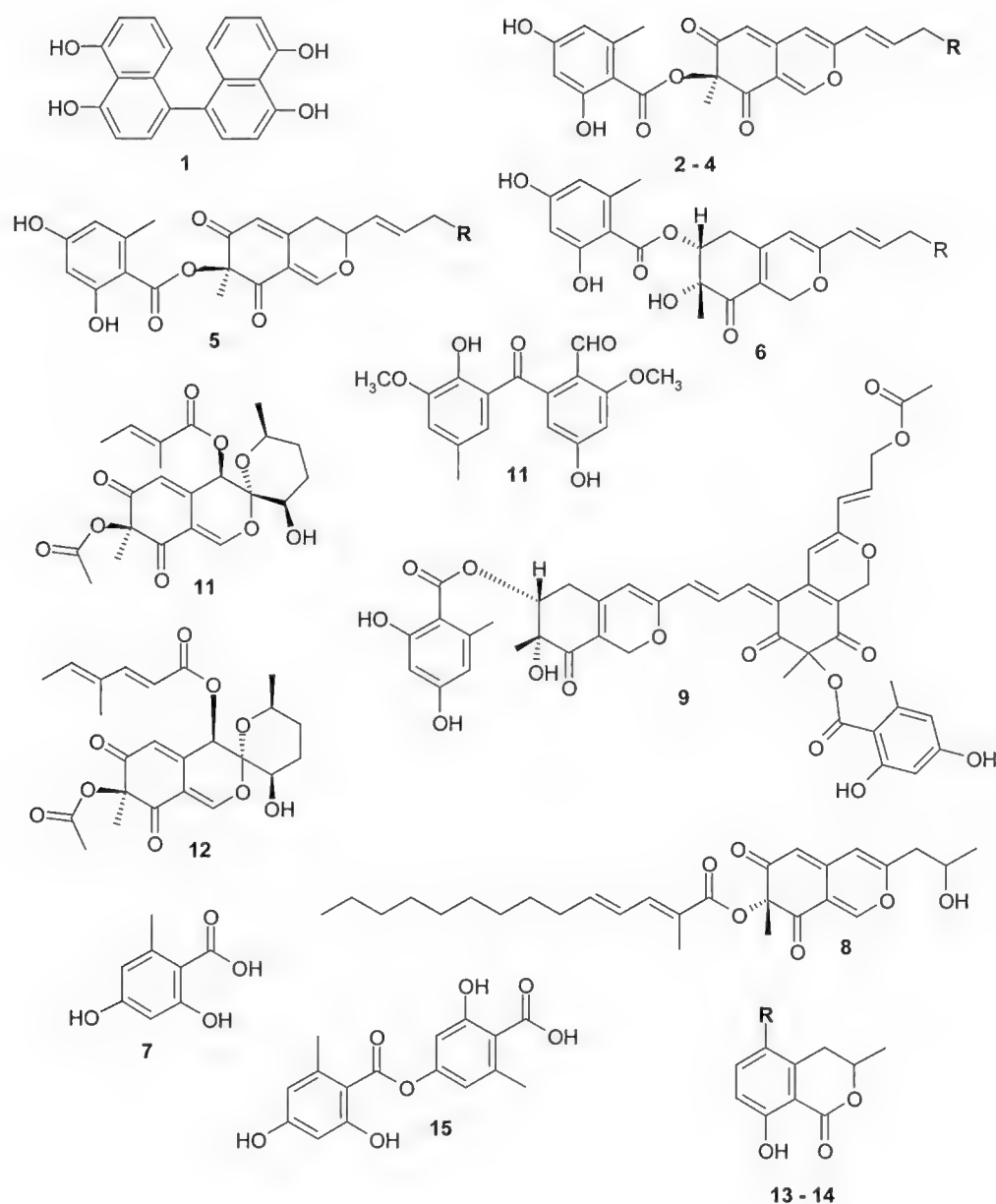


FIG. 1. Chemical structures of characteristic pigments and other secondary metabolites of *Hypoxylon* and allied genera, detected in this study by HPLC. 1: Binaphthalene tetrol (BNT); 2: Mitorubrin (R = H); 3: Mitorubrinol (R = OH); 4: Mitorubrinol acetate (R = OCCH<sub>3</sub>); 5: Hypomiltin (R = OCCH<sub>3</sub>); 6: Rubiginosin A (R = OCCH<sub>3</sub>); 7: Orsellinic acid; 8: Rubiginosin C; 9: Rutilin A; 10: Daldinal A; 11: Daldinin C; 12: Daldinin E; 13: Mellein (R = H) 14: 5-Methylmellein (R = CH<sub>3</sub>); 15: Lecanoric acid.

in conjunction with chemotaxonomy (i.e., stomatal pigment colors in 10% KOH). *Hypoxylon* was thus restricted to stomatic pyrenomycetes with an essentially homogenous stomatal context and *Nodulisporium*-like anamorphs. After erection of the genus *Annulohypoxylon* Y.M. Ju et al. (Hsieh et al. 2005) for sect. *Annulata* of *Hypoxylon* sensu Ju & Rogers (1996), *Hypoxylon* s. str. is now restricted to their sect. *Hypoxylon*.

In past years, we have studied several thousands of herbarium specimens and fresh material of *Hypoxylon* spp. from around the world. In addition to the characters deemed diagnostically important by Ju & Rogers (1996), we



studied secondary metabolite profiles recorded by high performance liquid chromatography coupled with diode array detection and mass spectrometry (HPLC-DAD/MS; cf. Hellwig et al. 2005, Stadler et al. 2001, 2004, 2008). Such HPLC profiles have proved quite valuable, because the production of secondary metabolites was largely found to be consistent in a given species, with the characteristic stromatal metabolites remaining stable even in ancient specimens collected up to 200 years previously. Due to this work, a comprehensive matrix of chemical and morphological data has become available that facilitates substantially the recognition of new taxa. On the other hand, novel biologically active compounds with potential utility were often encountered in rare species of *Hypoxylon*, as exemplified by the discoveries of rutilins (Quang et al. 2005) and carneic acids (Quang et al. 2006).

The current paper describes four new species of *Hypoxylon* from Western Europe and Eastern Africa that deviate significantly from all described taxa with respect to their morphological and chemotaxonomic traits.

### Materials and methods

Teleomorphic structures were microscopically observed in water (to study ascospore morphology), in Melzer's reagent (to test for amyloid ascal apical structures), in Chlorazol black (to measure ascal stipes), and in 10% KOH (to test for perispore dehiscence). In cases of apparent absence or lack of reactivity of ascal apical structures in Melzer's reagent, a pretreatment by 3% KOH was attempted. Ascospores were measured in water at 1000x magnification. KOH-extractable pigments were obtained as described in Ju & Rogers (1996). Color codes follow Rayner (1970). Ascospores were photographed in water or 10% KOH. Anamorphic structures were observed microscopically in water at 400–1000x magnification using phase contrast.

Cultures were obtained from ascospores prepared from perithecial contents on yeast-malt glucose (YMG) medium supplemented by antibiotics (Stadler et al. 2008). For morphological studies, the cultures were grown YMG and Difco Oatmeal agar (OA).

HPLC analyses of stromatal methanolic extracts were carried out according to Stadler et al. (2008) in two different gradients, using UV-visual detection (HPLC-UV/Vis) with diode array detection (DAD) and mass spectrometric detection (HPLC-MS) in both the positive and negative electrospray ionisation (ESI) mode. Secondary metabolites were identified by matching their retention times (Rt), HPLC-DAD and HPLC-ESI-MS spectra with external or internal standards of pure compounds that had been obtained previously. HPLC data of extracts and pure compounds from previous studies on *Xylariaceae* (Hellwig et al. 2005, Bitzer et al. 2007, Stadler et al. 2004, 2008) were also used for comparison. Cultures were propagated on YMG medium, and their extracts analyzed on the occurrence of secondary metabolites as described by Bitzer et al. (2008). Some compounds that were detected in the new taxa described herein or in morphologically similar species are depicted in FIG. 1. The trivial names of these compounds have been assigned a **bold** number in the legend of FIG. 1, to which they are referred in the taxonomic part.

***Hypoxylon fuscoides*** J. Fourn., P. Leroy, M. Stadler & Roy Anderson, **sp. nov.**

MYCOBANK MB 516748

FIGS 2–4

*A Hypoxylon fuscum differt granulis violaceis ad vinaceis in KOH dissolutis; a Hypoxylon rosieri differt ascosporibus ellipsoideo-inequilaterales, apicibus angustatis,  $9.5\text{--}12.5 \times 5\text{--}6 \mu\text{m}$ , riminibus germinativis sigmoideis praeditae. Status anamorphosis ad genero Virgariella similis.*

TYPE: FRANCE, VOSGES, Forêt de Rambervilliers, on bark of *Betula pendula* (Betulaceae), 7.X.2003, Paul Leroy, PL 03142B (HOLOTYPE – LIP; culture in MUCL 52670 and CBS 126418).

ETYMOLOGY: Latin, for its strong resemblance to *Hypoxylon fuscum*

STROMATA (FIG. 2) erumpent from bark, pulvinate, slightly constricted at base, gregarious, separate to coalescent, 1.4–3 mm diam  $\times$  0.8–1.4 mm thick; surface pruinose, Brown Vinaceous (84), pruina made up of red brown granules turning bluish green in 10% KOH, slightly uneven, with perithecial contours not exposed, with a thick layer of yellowish waxy granules beneath the surface turning colorless in 10% KOH, the whole stroma yielding Vinaceous Purple (101) pigments in 10% KOH; the tissue beneath the perithecia 0.5–1.2  $\mu\text{m}$  thick, greyish brown with blackish marks, soft-textured. PERITHECIA subglobose to obovoid, rarely slightly tubular, 0.32–0.38 mm high  $\times$  0.13–0.22 mm diam. OSTIOLES umbilicate, inconspicuous. ASCI (FIG. 3) cylindrical, short-stipitate, 8-spored, readily deliquescent, 100–120  $\mu\text{m}$  total length, the spore bearing-parts 70–84  $\mu\text{m}$  long  $\times$  7–8  $\mu\text{m}$  broad, the stipes 24–42  $\mu\text{m}$  long, with a discoid apical ring 0.5–0.8  $\mu\text{m}$  high  $\times$  3–3.4  $\mu\text{m}$  broad, bluing in Melzer's reagent. Paraphyses filiform, septate. ASCOSPORES (FIG. 3)  $9.5\text{--}12.5 \times 5\text{--}6 \mu\text{m}$  ( $M = 11 \times 5.4 \mu\text{m}$ ,  $n = 30$ ) ellipsoid slightly inequilateral with narrowly rounded to acute ends, brown, smooth, with a conspicuous spore-length sigmoid germ slit, swelling rapidly in water; perispore dehiscent in 10% KOH, thin-walled, with faint transverse striae. Epispore smooth.

CULTURES AND ANAMORPH: COLONIES on OA covering Petri dish in 2–3 weeks, at first white, becoming Hazel (88), velvety, azonate, with diffuse margins; reverse remaining uncolored. Sporulating regions in patches, vinaceous buff (86). Conidiogenous structure referable to the *Virgariella*-like branching pattern as defined by Ju & Rogers (1996), hyaline, smooth to finely roughened. CONIDIOGENOUS CELLS (FIG. 4) hyaline, smooth,  $8\text{--}14(-25) \times 2.5\text{--}4 \mu\text{m}$ , often arranged repetitively at the tips of the conidiophores (Fig. 4), so that up to ten conidiogenous cells are produced in succession. CONIDIA hyaline, smooth, ellipsoid,  $5\text{--}7 \times 2.5\text{--}3 \mu\text{m}$ .

SECONDARY METABOLITES: HPLC profiling (FIG. 5A) revealed that this species differs from all morphochemotypes of *H. fuscum*, we have hitherto studied, regardless from which host plants their stromata have been encountered, in lacking daldinins C, E and F and daldinal A. The lack of these pigments clearly

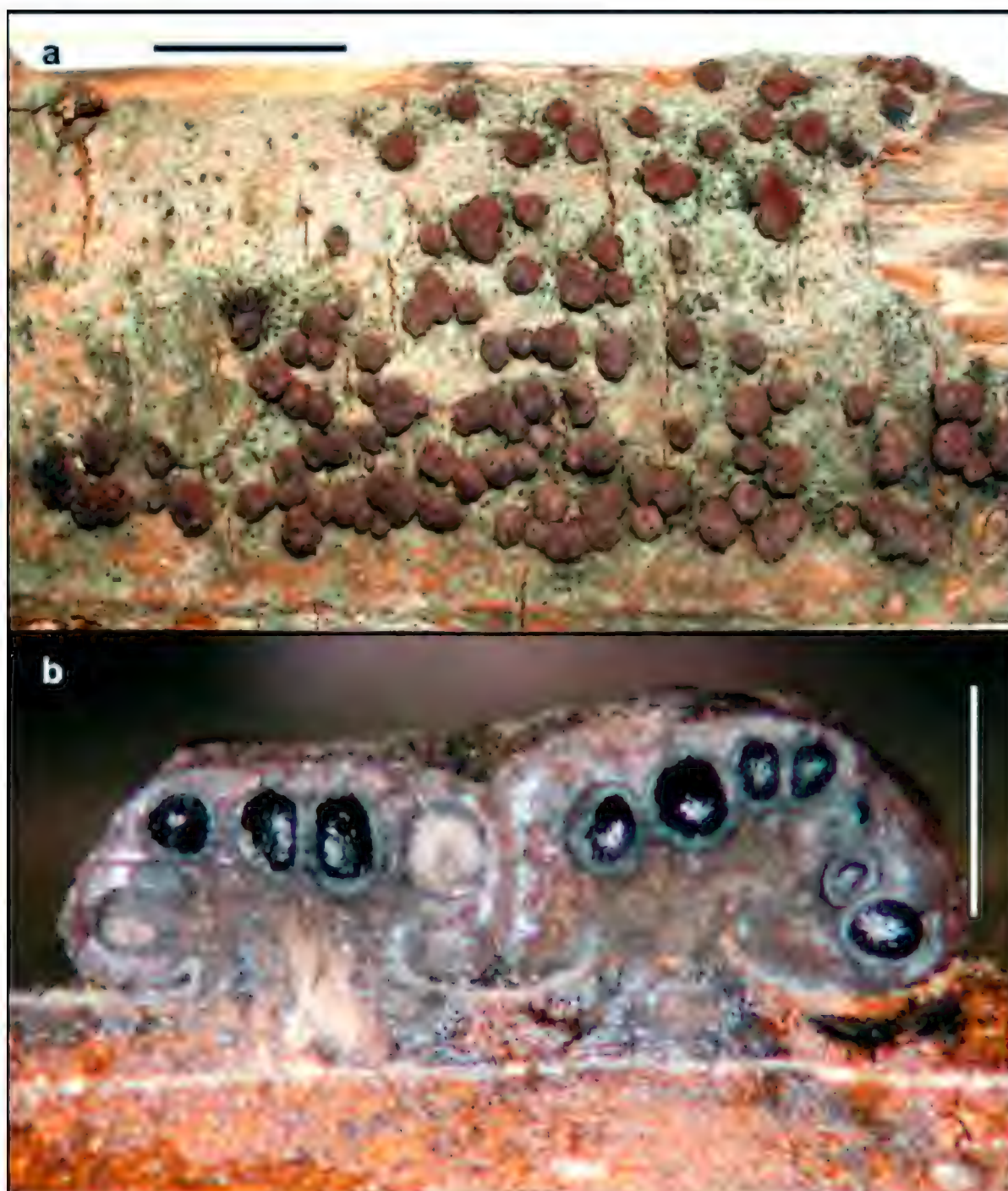


FIG. 2. Stromata of *Hypoxylon fuscoides*, from holotype (PL 03142B). a. Stromatal habit on the natural substrate. b. Section through stroma, showing the ruptured periderm and perithecial arrangement. Scale bars: a. 1cm, b: 1 mm.

accounts for the pigments in KOH being purple, rather than olivaceous brown. Binaphthalenes (in particular binaphthalene tetrol, BNT) were found to be the prevailing stromatal metabolites. The cultures produced 5-methylmellein (14) as major component in YMG medium.

FURTHER SPECIMENS EXAMINED: UNITED KINGDOM, NORTHERN IRELAND. Vice-county H37 (Armagh), OXFORD ISLAND (J045620), on bark of fallen branches of *Alnus incana* (Betulaceae), 18.X.2007. R. Anderson. Vice-county H38 (Down), BELFAST, Belvoir Forest, (J333693), on bark of fallen branches of *Alnus incana*, 5.II.2008. R. Anderson (K, culture in MUCL 52423).



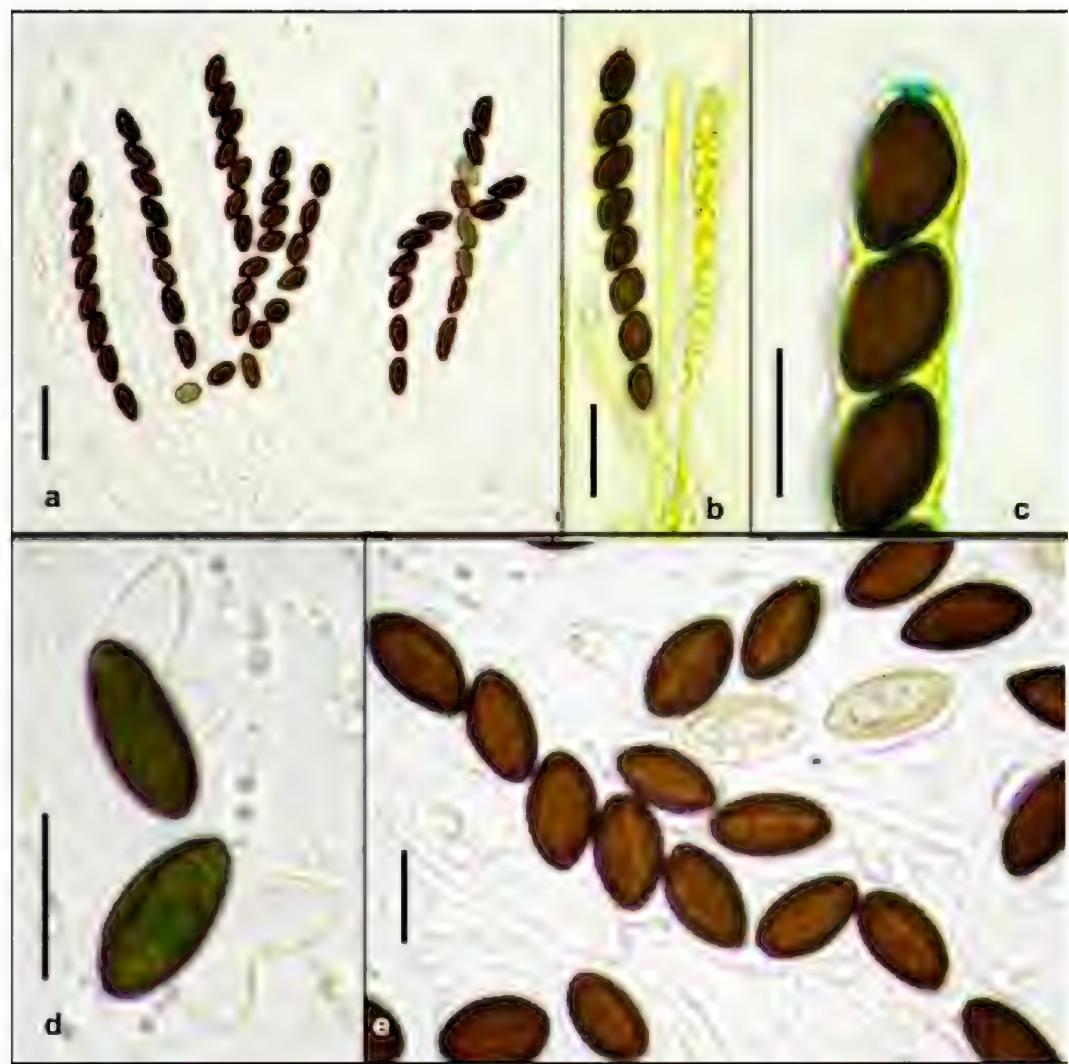


FIG. 3. Microscopic characteristics of *Hypoxylon fuscoides*, from paratype (JF 09347; K). a. Asci in water. b, c. Asci in Melzer's reagent, c showing amyloid apical apparatus. d Ascospores in KOH, showing dehiscent perispore. e. Ascospores in water, showing the sigmoid germ slits. Scale bars: a, b: 20  $\mu\text{m}$ , c, d, e: 10  $\mu\text{m}$

COMMENTS: *Hypoxylon fuscoides*, already mentioned on a website dedicated to fungal taxonomy (Fournier & Magni 2004) and by Anderson (2008), is not distinguishable from *H. fuscum* in the field. Despite the fact that these species share many morphological features, the new taxon can be readily separated by its reaction in KOH and its smaller ascospores with more acute ends.

*Hypoxylon fuscum* as currently conceived (Petrini & Müller 1986, Ju & Rogers 1996, Granmo 1999) features a very wide ascospore size range that is not clearly correlated with other morphological, ecological, or chemotaxonomic characters. In the present case, the deviating morphology of ascospore ends appears more significant than the difference in size. The holotype collection has paler pigments in KOH than the material from Northern Ireland, but the ascospores are identical, and it features similar very small gregarious stromata. The cultures are similar to those described for *H. fuscum* by Petrini & Müller (1986) and Ju & Rogers (1996) but have rather stout conidiogenous cells and



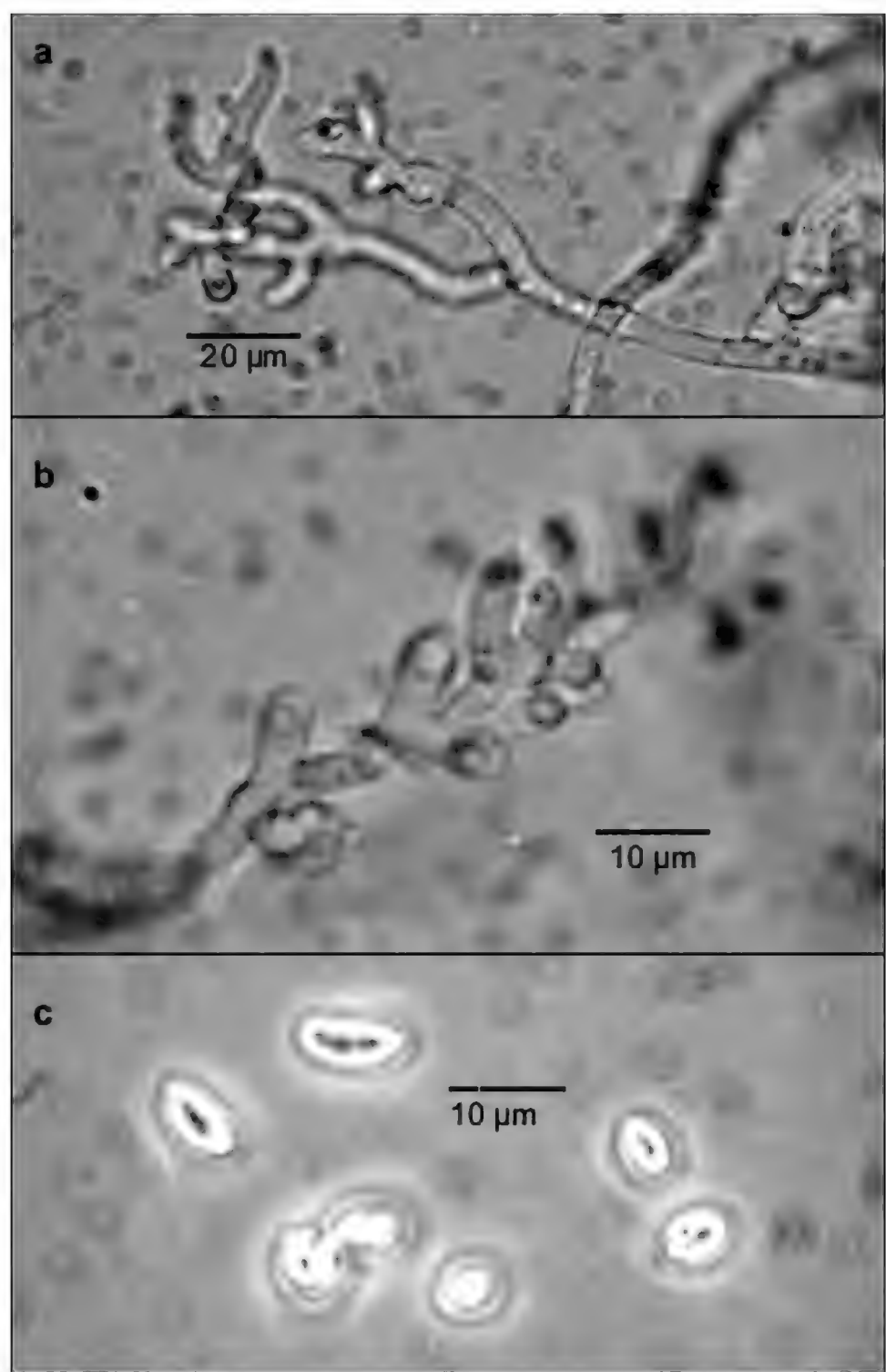


FIG. 4. *Hypoxylon fuscoides*, ex-type strain, from OA culture. a. conidiophores, showing dichotomously branched *Virgariella*-like conidiogenous structures. b. Close-up of conidiophore apex, showing repetitive branching, resulting in stout conidiogenous cells. c. Conidia. Scale is indicated by bars.

slightly larger conidia. In particular, the successive production of numerous small conidiogenous cells from the tip of the same conidiophore is only exceptionally observed in other cultures of *Hypoxylon*, and those of the most frequent morphochemotype of *H. fuscum* from *Corylus* normally produce conidiogenous cells up to 40 µm long. The stromatal HPLC profile also deviates strongly from that observed in numerous collections of *H. fuscum* collected

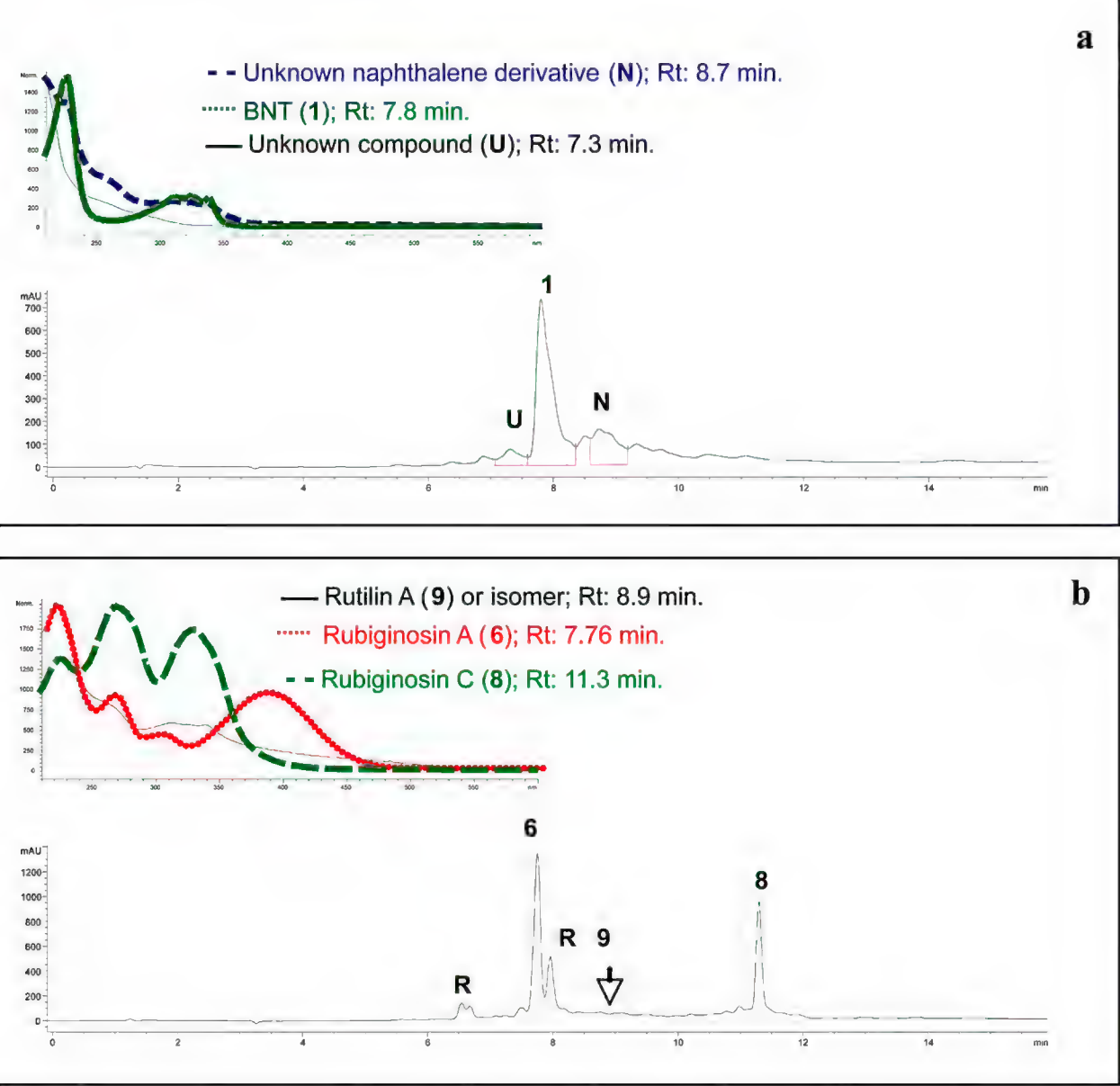


FIG. 5. Stromatal HPLC-UV profiles (210 nm) of holotype specimens of *Hypoxylon fuscoides* (a) and *Hypoxylon lusitanicum* (b). and DAD spectra of major metabolites. In FIG 5a, U indicates an unknown major component lacking a characteristic DAD spectrum, and N indicates an unknown naphthalene with a DAD spectrum similar to BNT (1). Daldinal A and daldinins C and E (10 - 12), the characteristic pigments of *H. fuscum*, were not detected. In FIG. 5b, R indicates further major components of the rubiginosin type, whose spectra are not depicted). For chemical structures of known compounds see FIG. 1.

from *Alnus* and *Salicaceae*, which apparently also lacks daldinins but contains different pigments that also result in olivaceous colors in KOH in our recent study (Stadler et al. 2008).

Rogers et al. (2008) described *H. rosieri* J.D. Rogers & Lar.N. Vassiljeva, another segregate of *H. fuscum* from the USA (Texas), based on a similar purple KOH reaction. Their species differs from *H. fuscoides* in having markedly longer, more slender ascospores ( $13.5\text{--}15 \times 5\text{--}6 \mu\text{m}$ ). Furthermore, sigmoid ascospore germ slits were not mentioned in their description.

***Hypoxylon lusitanicum* J. Fourn., M. Stadler & Priou, sp. nov.**

FIGS 6–7

MYCOBANK MB 516749

*A Hypoxylon perforato differt granulis rufobrunneis in KOH dissolutis et ascis longistipitatis, 158–170 µm longitudine tota, partibus sporiferis 76–91 × 7–8 µm, stipitibus 70–80 µm.*

TYPE: PORTUGAL: RIBATEJO Prov., Achete, RIBEIRHINA, 39° 19' 13" N, 08° 42' 55" W, alt 55 m., on dead blackened wood of *Rhamnus alaternus* (*Rhamnaceae*) in Mediterranean evergreen vegetation, 5.V.2009, J.P. Priou, JF 09125 (HOLOTYPE – LIP; ex-type culture in MUCL52424).

ETYMOLOGY: For Portugal (Lusitania in Latin).

STROMATA (FIG. 6) effused, ellipsoid to elongated, 8–22 mm long × 2.5–8 mm broad × 0.6–0.8 mm thick, at times coalescent, at times with steep, indented black margins; surface pruinose, slightly uneven, Brown Vinaceous (84), with perithecial contours hardly exposed, with a thick layer of olivaceous yellow waxy granules beneath the surface and around the upper half of perithecia, yielding Sienna (8) pigments in 10% KOH; the tissue beneath the perithecia 50–150 µm thick, dull brown, soft-textured, delimited by a black line spreading over the underlying wood. PERITHECIA subglobose to obovoid, 0.5–0.6 mm high × 0.3–0.45 mm diam. OSTIOLES umbilicate, often in a shallow depression, fringed with a disc of white material 70–80 µm diam. ASCI (FIG. 7) cylindrical, long-stipitate, 8-spored, 158–170 µm total length, spore bearing-parts 76–91 µm long × 7–8 µm broad, the stipes 70–80 µm long, with a discoid apical ring 0.8–1 µm high × 2.5–3 µm broad, bluing in Melzer's reagent. Paraphyses not seen. ASCOSPORES (FIG. 7) 11–13.5 × 5–7 µm ( $M = 11.8 \times 5.5 \mu\text{m}$ ,  $n = 30$ ) ellipsoid-inequilateral with narrowly rounded to acute ends, brown, smooth, with a spore-length straight germ slit; perispore readily dehiscent in 10% KOH, faintly striate. Episore smooth.

CULTURES AND ANAMORPH: Colonies on YMG and Difco OA covering Petri dish in 2 weeks, at first whitish, becoming umber (9), velvety to felty, azonate, with diffuse margins, with honey (64) pigments diffused beyond colonies; reverse slightly melanizing with age. No conidiogenous structures observed.

SECONDARY METABOLITES: In accordance with the orange pigments in KOH, the stromatal HPLC profile of *H. lusitanicum* (Fig. 5b) revealed the presence of azaphilones, with rubiginosins A (6) and C (8) being major detectable components. A minor metabolite at Rt 8.9 min was also observed, which probably corresponds with rutilin A (9) or another yet unknown dimeric azaphilone of the rutilin type. Neither mitorubins nor hypomiltin (2 – 5) were detected. However, the cultures produced 5-methylmellein (14) as major component in YMG medium, indicating a relationship to the *H. fuscum* and *H. rubiginosum* species complexes (cf. Bitzer et al. 2008).

FURTHER SPECIMEN EXAMINED: PORTUGAL: RIBATEJO Prov., Achete, RIBEIRHINA, *Rhamnus alaternus*, 6.V.2009, mixed with old pulvinate stromata of *Hypoxylon perforatum*, J.P. Priou, JPP 29083.



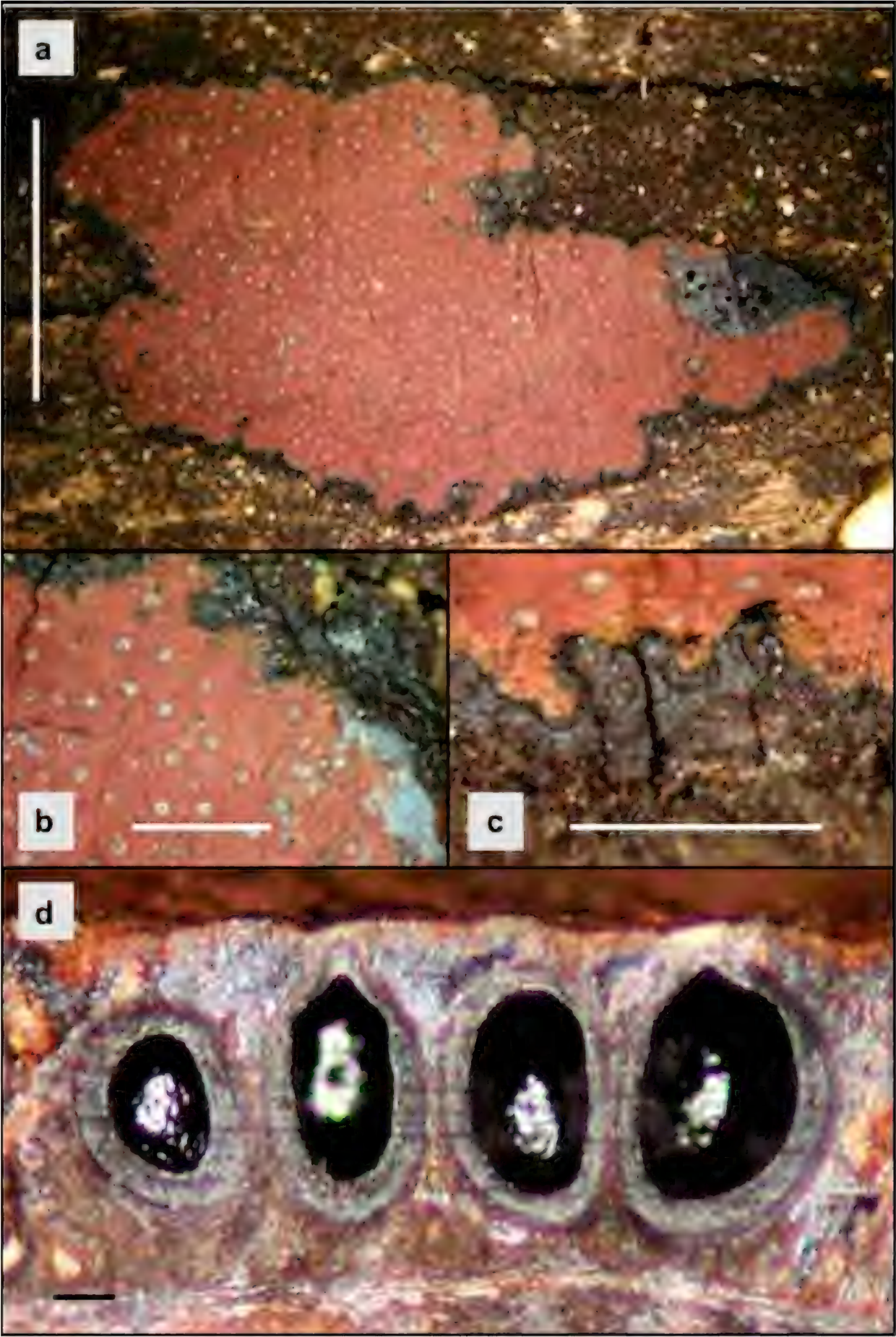


FIG. 6. Stromata of *Hypoxylon lusitanicum*, from holotype (LIP). a. Stromatal habit on the natural substrate. b, e. Close-up of stromatal surface, showing ostioles. c. Close-up of blackened stromatal margin. d. Section through stroma, showing perithecia. Scale bars: a: 5 mm, b, c: 1mm d: 0.1 mm.



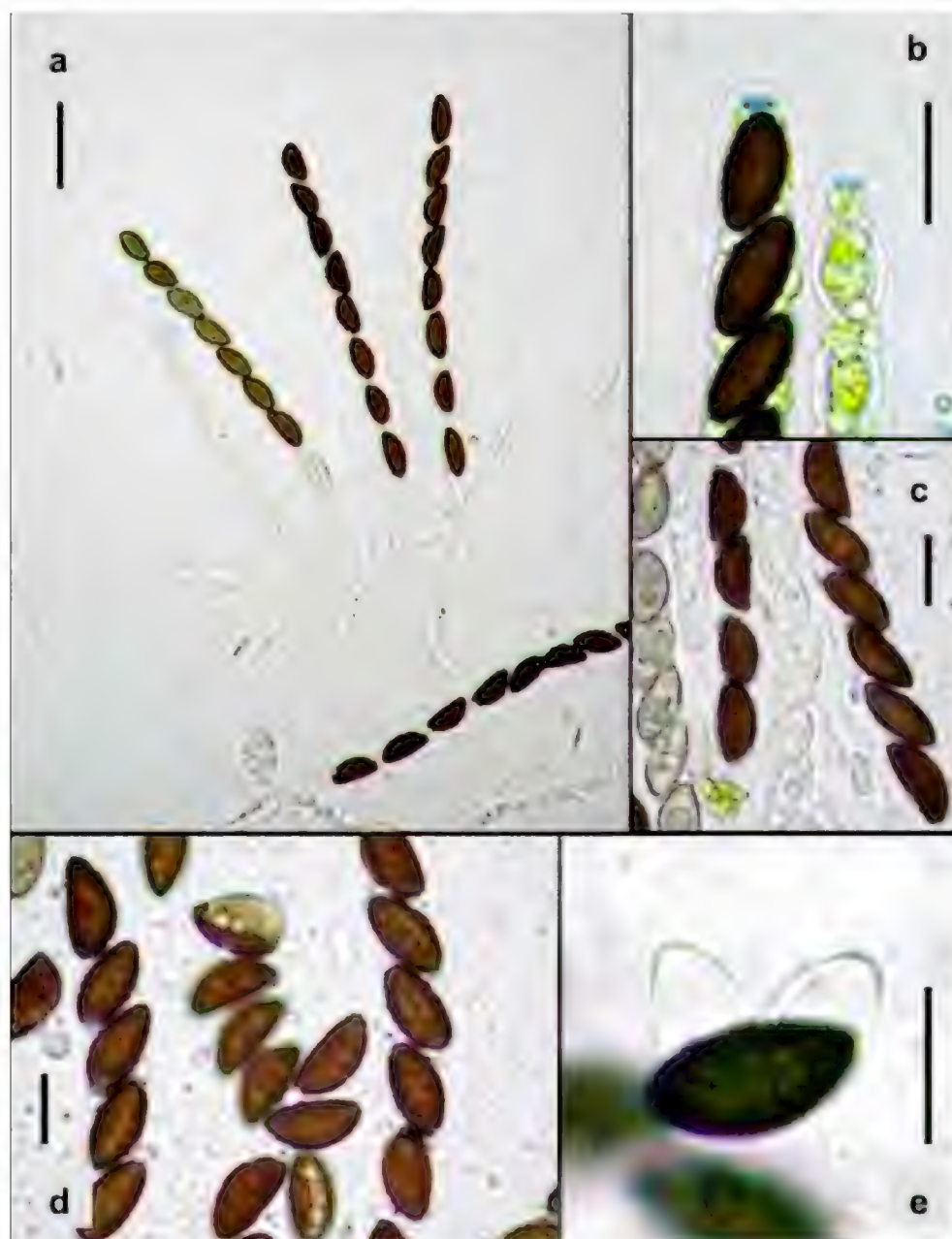


FIG. 7. Microscopic characteristics of *Hypoxylon lusitanicum*, from holotype (LIP). a, Asci in water. b Ascus tip in Melzer's reagent, showing amyloid apical apparatus. c-d Ascospores in water. e Ascospore in KOH, showing dehiscent perispore. Scale bars: a: 20 µm, b, c, d, e: 10 µm.

COMMENTS: *Hypoxylon lusitanicum* appears highly similar to *H. perforatum* with regard to its stromatal morphology (conspicuous white discs around umbilicate ostioles and presence of yellowish granules beneath the stromatal surface). It can be distinguished by its red brown pigments in KOH, larger perithecia, long-stipitate asci, and significantly larger ascospores with more narrowly rounded ends. The red brown pigments are due to the presence of rubiginosins as in *H. rubiginosum* and *H. petriniae*, whereas *H. perforatum* produces hypomiltin instead (cf. Stadler et al. 2004). Both, *H. rubiginosum* and *H. petriniae* have often been confused with *H. perforatum*; hence they might be easily confounded with *H. lusitanicum*. Two recently described taxa from the Canary Islands, *H. canariense* and *H. urriesii* (Stadler et al. 2008), might also

TABLE 1. Diagnostic characters of six species of the *Hypoxylon rubiginosum* complex.

	<i>H. canariense</i>	<i>H. lusitanicum</i>	<i>H. perforatum</i>	<i>H. petriniae</i>	<i>H. rubiginosum</i>	<i>H. urriesii</i>
SURFACE COLOR	Fulvous(43), Dark Brick (60), or Brown Vinaceous (84)	Brown Vinac. (84)	Dark Brick (60) to Brown Vinac. (84)	Vinaceous Grey (116) to Brown Vinac. (84)	Rust ( 39) to Dark Brick (60)	Dark Brick (60)
WHITE- FRINGED OSTIOLES	frequent	present	present	frequent	occasional	absent
STROMAL THICKNESS	0.5–0.6 mm	0.6–0.8 mm	0.5–1–(2.5) mm	0.3–0.8 mm	1–1.3(–2) mm	0.3 mm
PERITHECIAL DIAM.	0.3–0.4 mm	0.3–0.45 mm	0.25–0.4 mm	0.25–0.4 mm	0.3–0.65 mm	0.15–0.2 mm
KOH PIGMENTS	Orange(7) to ienna(8)	Sienna (8)	Amber (47)	Orange (7)	Orange (7)	Orange (7)
SECONDARY METABOLITES	mitorubrins, rubiginosins	rubiginosins, rutilin A	hypomiltin	rubiginosins, BNT	mitorubrins, rubiginosins	mitorubrins,r ubiginosins
ASCUS STIPE LENGTH	27–40 µm	70–80 µm	24–50 µm	37–64 µm	60–98 µm	18–30 µm
AV. ASCOSPORE SIZE	10.4 × 4.8 µm	11.8 × 5.5 µm	10.9 × 4.9 µm	10.7 × 5.1 µm	10.1 × 4.4 µm	12.3 × 5.4 µm
GERM SLIT	straight	straight	straight	straight	straight	sigmoid
PERISPORE (LM)	smooth	faintly striate	smooth	smooth	smooth	smooth
REFERENCE	Stadler et al. 2008	This study	Fournier & Magni 2004	Fournier & Magni 2004	Fournier & Magni 2004	Stadler et al. 2008

be confused in the field with *H. lusitanicum* because of their effused stromata having similar surface colors. *Hypoxylon canariense* mainly differs from *H. lusitanicum* in having short-stipitate asci and smaller ascospores averaging  $10.4 \times 4.8 \mu\text{m}$  with a smooth perispore, while *H. urriesii* differs from the new taxon in having much smaller perithecia, short-stipitate asci and slightly larger ascospores averaging  $12.3 \times 5.4 \mu\text{m}$  with a sigmoid germ slit and a smooth perispore. Some important diagnostic characters to discriminate these six species are summarized in TABLE 1.

***Hypoxylon gibriacense* J. Fourn., M. Stadler & Gardiennet, sp. nov.**

MYCOBANK MB 516750

FIGS 8–9

*A Hypoxylon shearii et Hypoxylon fraxinophili differt discis annulatis conspicuis, peritheciis ambientibus. A Hypoxylon addis differt in ascosporae parviorae, perisporium conspicuiter striatum praeditae.*

TYPE: FRANCE, CÔTE D'OR, Gevrey-Chambertin, COMBE DE LAVAUX, on moss-covered bark of a fallen branch of *Acer platanoides* (Aceraceae), 3.XII.2009, A. Gardiennet, AG 09033 (HOLOTYPE – LIP; culture in MUCL 52698).

ETYMOLOGY: from *Gibriacum*, the Latin name of Gevrey, the locality of the type collection.

STROMATA (FIG. 8) corticolous, erumpent through the periderm, glomerate with a narrowly restricted base, containing 12–20 perithecia, scattered to often coalescent, 2–3 mm diam  $\times$  1.5–2.5 mm thick, soft-textured; surface Greyish Sepia (106) with a faint olivaceous tone, pruinose, with perithecial contours exposed to strongly exposed; dull yellow granules forming a thin crust beneath the surface and sometimes extending between the perithecia, yielding fugacious Amber (47) then Sienna (8) pigments in 10% KOH (Livid Red (56) under the microscope); subperithecial tissue purplish black, brownish grey at base 1–1.3 mm thick. PERITHECIA ellipsoid to subglobose, 0.5–0.6 mm high  $\times$  0.4–0.5 mm diam. OSTIOLES umbilicate, opening at the centre of a paler discoid area ca. 0.2 mm diam delimited by a low rim. ASCI (FIG. 8) unitunicate, cylindrical, 130–140  $\mu\text{m}$  total length, the spore-bearing parts 85–95  $\mu\text{m}$  long  $\times$  9–9.5  $\mu\text{m}$  broad, the stipes 40–45  $\mu\text{m}$  long, apex without apical ring, not bluing in Melzer's reagent. Paraphyses filiform, copious. ASCOSPORES (FIG. 9) 11.5–13  $\times$  6–6.8  $\mu\text{m}$  ( $M = 12.4 \times 6.5 \mu\text{m}$ ,  $n = 30$ ), ellipsoid-inequilateral with narrowly rounded ends, one side flattened to sometimes slightly concave, brown to dark brown, smooth, with spore-length straight germ slit (arrows). Perispore dehiscent in KOH, with fairly conspicuous striae somewhat anastomosing. Epispore smooth.

CULTURES on YMG and OA media covering a 9 cm Petri dish in 2–3 weeks, white, felty to floccose, azonate, with diffuse margins; reverse becoming Honey (64). No conidiophores or other anamorphic structures observed after up to 6 weeks of incubation.



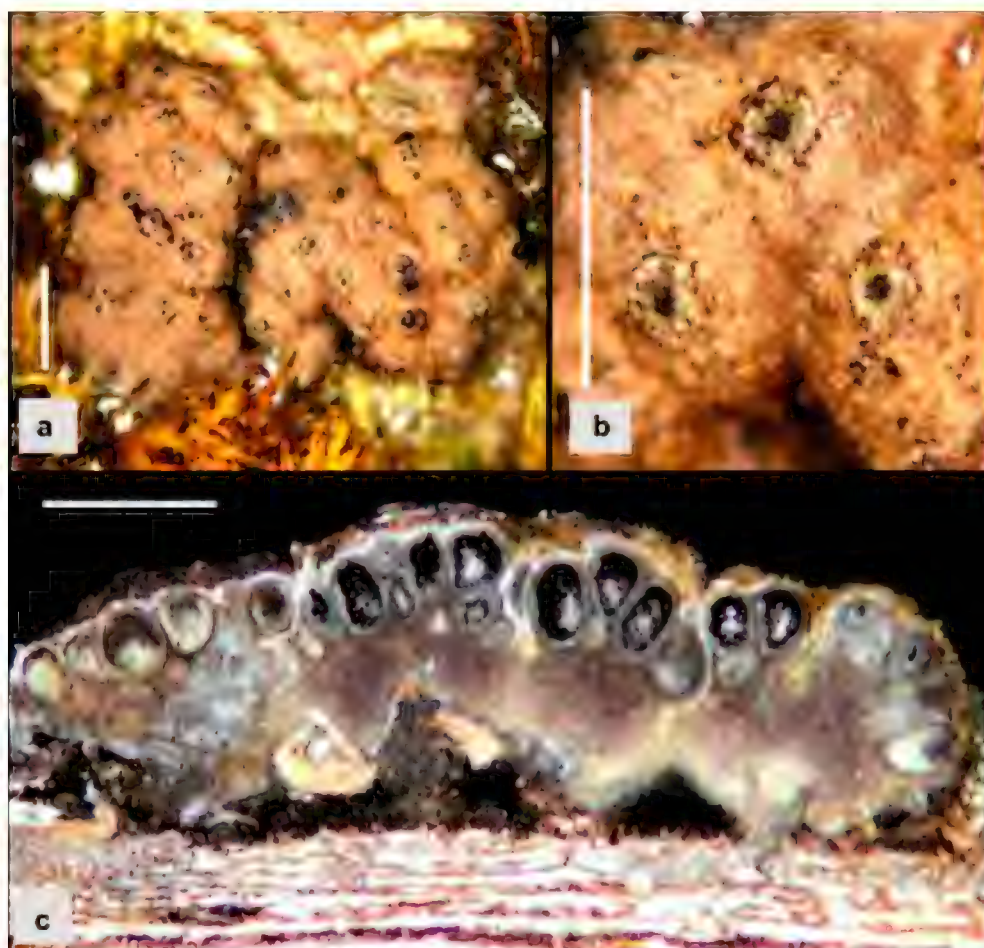


FIG. 8. Stromata of *Hypoxylon gibriacense*, from holotype (LIP). a. Stromatal habit on the natural substrate. b. Close-up of stromatal surface, showing perithecial mounds and ostiolar disks. c. Section through stroma, showing perithecia. Scale bars: a, b, c: 1 mm.

SECONDARY METABOLITES: HPLC of the stromatal MeOH extract of the holotype specimen (FIG. 13) revealed rubiginosin C (6) and another major peak that was revealed to be a mixture of BNT (1) and hypomiltin (5) only by HPLC-MS because the chromatographic method used to separate the components in the crude extract by HPLC-DAD appeared insufficient to discriminate these compounds. The DAD spectrum therefore at first appeared unique because it was actually caused by two components showing different absorption maxima in the UV-visual detection range. The mass spectra derived from this peak, labeled (“1 + 5”) containing both compounds are included in FIG. 13 for comparison. All these compounds also occur in various other species of the *H. rubiginosum* complex (cf. Stadler et al. 2008). The cultures (FIG. 15) produced mellein (13) and several other metabolites, most of which were not yet identified, but 5-methylmellein (14) was not observed.

COMMENTS: *Hypoxylon gibriacense* is distinctive in featuring ostiolar discs and in having asci lacking an apical apparatus. Despite the clearly differentiated discs around the ostioles, it is considered best placed in *Hypoxylon* rather than *Annulohypoxylon* based on the soft-textured stromata and ascospores with transversally dehiscent and striate perispores that lack a dorsal thickening



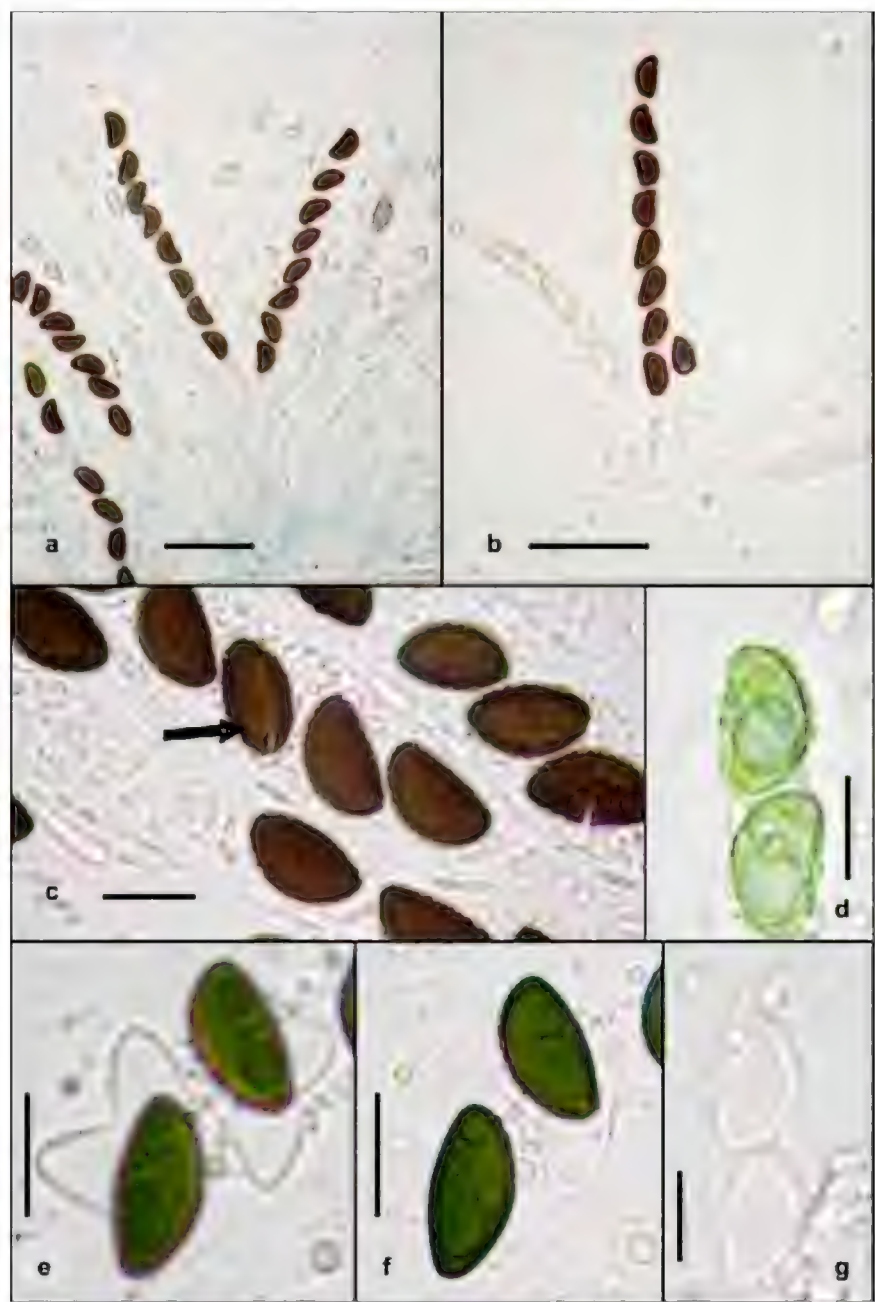


FIG. 9. Microscopic characteristics of *Hypoxylon gibriacense*, from holotype (LIP). a, Asci in chlorazol black. b, ascus in water. c Ascospores in water. d. Ascus tip in Melzer's reagent e, f. Ascospores in KOH at different focuses, showing dehiscence ornamented perispores. g. Free perispores. Scale bars: a b.: 30 µm, c- g: 10 µm

(FIG. 9). The inconspicuous glomerate stromata with conspicuous perithecial elevations and thick subperithecial tissue, the presence of yellow granules beneath the surface yielding red brown pigments in KOH, the lack of ascial apical rings, and the conspicuously striate perispores make a combination of characters not known from any temperate or tropical taxon of *Hypoxylon*.

Of the known European (and Northern temperate) taxa, *H. fraxinophilum* (Pouzar 1972) appears most similar. The stromatal, ascial, and ascospore morphology of this fungus are reminiscent of *H. gibriacense*. As previously shown by Stadler et al. (2004, as "*H. intermedium*"), *H. fraxinophilum* also contains hypomiltin (5), but neither rubiginosin C (6) nor BNT (1) were

detected in its stromata. Furthermore, it differs in its stromata lacking ostiolar disks, and in its host specificity for *Fraxinus*, rather than *Acer*. The new species is, however, remarkably similar to *H. addis* (see below) in having ostiolar discs, similar pigment colors in KOH, and its asci lacking an amyloid apical apparatus. *Hypoxylon gibriacense* and *H. addis* differ in their ascospore dimensions and in the merely faintly striate perispores in *H. addis*; moreover, the stromatal secondary metabolite profiles of the two species differ completely.

***Hypoxylon addis*** J. Fourn., M. Stadler & U. Lindem., **sp. nov.**

MYCOBANK MB 516751

FIGS 10–11

*A Hypoxylon shearii et Hypoxylon fraxinophili differt discis annulatis conspicuis, peritheciis ambientibus. A Hypoxylon gibriacense differt in ascosporae maiora.*

TYPE: ETHIOPIA: GIYON/WOLISSO, Negash Lodge, 2000m, +8° 32' 1.73", +37° 58' 52.65", on a corticated dry twig of *Croton sylvaticus* (Euphorbiaceae). 3.X.2009, U. Lindemann, JF-09302 (HOLOTYPE– LIP).

ETYMOLOGY: Ethiopian “Addis”, meaning “new”.

STROMATA (FIG. 10) corticolous, scattered, glomerate-pulvinate, erumpent through the periderm, 1–3 mm diam × 1–1.2 mm thick, soft-textured; surface Vinaceous Buff (86) to dark Brick (60), pruinose, with perithecial contours exposed to strongly exposed, at times rosellinoid; dull yellow granules beneath the surface and between the perithecia yielding Luteous (12) to Orange (7) pigments in 10% KOH; subperithecial tissue brownish with black streaks, 0.3–0.5 mm thick. PERITHECIA subglobose, 0.5–0.55 mm diam. OSTIOLES umbilicate, most often opening at the centre of a raised disc ca. 0.35 mm diam. ASCI (FIG. 11) unitunicate, cylindrical, 170–190 µm total length × 9.5–10.5 µm broad, the spore-bearing parts 85–100 µm long, the stipes 70–90 µm long, easily broken, apex without apical ring, not bluing in Melzer's reagent. Paraphyses filiform, copious. ASCOSPORES (FIG. 11) 13–16.5 × 6–7.7 µm ( $M = 14.6 \times 7 \mu\text{m}$ ,  $n = 20$ ), ellipsoid-inequilateral with narrowly rounded ends, one side flattened to often slightly concave, dark to blackish brown, smooth, with spore-length straight germ slit. Perispore dehiscent in KOH, striate, with striae visible in brightfield microscopy but inconspicuous, epispore smooth.

No cultures obtained. Anamorph not seen.

SECONDARY METABOLITES: Surprisingly, the HPLC profile of the stromata of *H. addis* did not reveal any known metabolites of *Hypoxylon* or other *Xylariaceae* that we have characterized or observed in the past. As shown in FIG. 14, the stromatal extract contained a predominant peak with a rather characteristic chromophore. Another, presumably related, minor component showing a highly similar DAD spectrum was observed at a lower Rt. A search in the HPLC library used for dereplication of natural products in crude extracts that represents several thousands of pure compounds (Bitzer et al 2007) revealed



that the prevailing stromatal metabolite of *H. addis* corresponds to lecanoric acid (16). The DAD and MS spectra and the Rt of lecanoric acid (16) are depicted in FIG. 14.

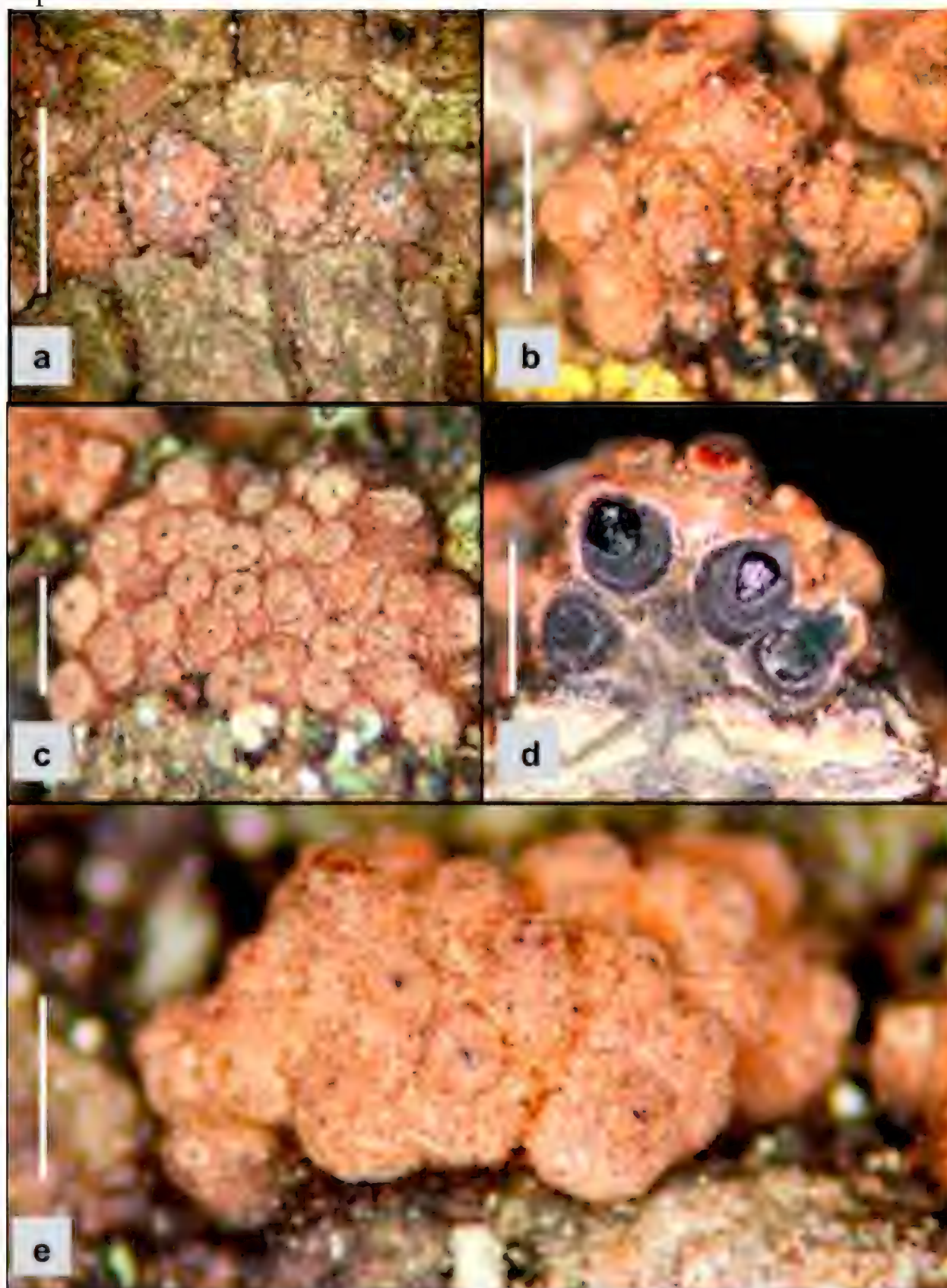


FIG. 10. Stromata of *Hypoxylon addis*, from holotype (LIP). a. Stromatal habit on the natural substrate. b, e. Close-up of stromatal surface, showing rosellinioid perithecial mounds. c. Surface of a glomerate stroma, showing characteristic ostiolar disks. d. Section through stroma, showing perithecia. Scale bars: a. 5 mm, b. 0.5 mm, c, d, e. 1 mm.

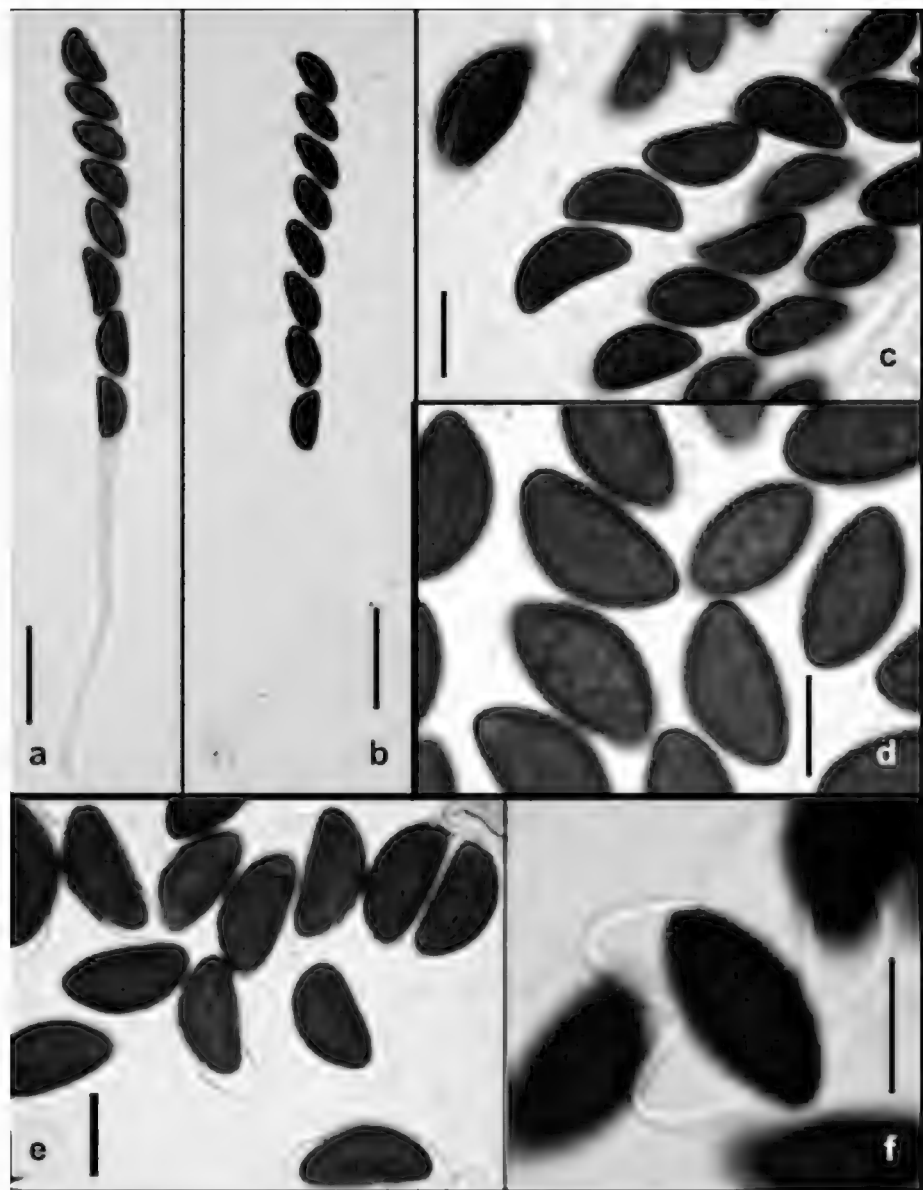


FIG. 11. Microscopic characteristics of *Hypoxylon addis* from holotype (LIP). a, b Asci in chlorazol black. c Ascospores in water. d-f Ascospores in KOH, showing germ slit (d) and dehiscent perispore. Scale bars: a, b: 20 µm, c, d, e, f: 10 µm.

MATERIAL STUDIED FOR COMPARISON (Fig. 12): USA: LOUISIANA, East Baton Rouge Parish, corticated wood of *Quercus*, IV.1980, J.D. Rogers & J.P. Jones (WSP 69637 – holotype of *H. shearii*).

COMMENTS: *Hypoxylon addis* is distinctive in its small glomerate stromata with large discoid ostioles and microscopically in its asci lacking an apical ring and rather large, dark-colored ascospores with a faintly striate perispore. The collector stated that he also found this species on a dry twig of *Cordia africana*, but that specimen was moldy and needed to be discarded. From a comparison of teleomorphic characters, *H. shearii* Y.M. Ju & J.D. Rogers (Ju & Rogers 1996) appears most similar with respect to its stromatal and ascospore morphology and the color of its stromatal pigments. The type specimen of *H. shearii* was studied for comparison (FIG. 12) and as previously reported (Stadler et al. 2008), its HPLC profile revealed mitorubins as well as





FIG. 12. Morphological characteristics of *Hypoxylon shearii*, from holotype (WSP). a. Stromata. b. Sectioned stromata showing yellow granules and globose perithecia. c. Ascospores in water. d. Ascospores in KOH, showing dehiscent perispore. Scale bars: a,b: 1 mm, c, d: 10 µm.

rubiginosins, all of which are absent in *H. addis*. The new species also differs in having conspicuous raised discs around the ostioles and microscopically in having larger ascospores ( $13\text{--}16.5 \times 6\text{--}7.7$  µm vs.  $12\text{--}14 \times 5.5\text{--}6.5$  µm in *H. shearii*) with much less conspicuous ornamentation on the perispore. We have not yet studied authentic material of *H. shearii* var. *minor* F. San Martín et al. (1999), which differs from the typical variety by having smaller ascospores,  $7\text{--}8 \times 3.5\text{--}4$  µm. Interestingly, both varieties of *H. shearii* have been collected thus far exclusively from *Quercus*.

One of the most intriguing features encountered in *H. addis* is the stromatal pigment profile, almost exclusively revealing lecanoric acid. This molecule is widely distributed in lichenized ascomycetes (Huneck 2001 and references cited therein) but has so far not often been encountered in non-lichenized fungi. According to our knowledge, the present study reveals lecanoric acid from a member of the *Xylariaceae* for the first time. Our retrospective analysis of the previously recorded HPLC profiling data in our *Xylariaceae* metabolite library confirms that lecanoric acid has indeed not been detected as major component of any of the previously studied 3500 specimens of *Hypoxylon*, including the majority of currently accepted taxa and their type specimens.

Lecanoric acid is formally derived from condensation of two molecules of orsellinic acid (7), which is widespread in the *H. rubiginosum* complex

as well as in the *H. fragiforme* group (Stadler et al. 2008). All the ubiquitous molecules of the mitorubrin, rubiginosin, and hypomiltin azaphilones contain an orsellinic acid moiety, attached to the azaphilone core molecules by an ester bond. Accordingly, the free orsellinic acid was found in many of the corresponding stromatal extracts of the respective *Hypoxylon* spp. as major component. The azaphilone core moieties were not detected in *H. addis*, although its stromata showed similar pigment colors in KOH as many species of the *H. rubiginosum* complex. Therefore, *H. addis* might represent a rather derived member of *Hypoxylon*, which has early abandoned or never attained azaphilone biosynthesis and developed the specific pathway for lecanoric acid instead, in convergence to the *Lecanorales* and other lichenized taxa of *Ascomycota*. It should be interesting to compare this species using molecular phylogenetic data in order to assess its closest relatives. However, we have so far been unable to obtain viable cultures from the stromata.

Although lichenized ascomycetes have been studied intensively for secondary metabolites for over a century, with many taxa of *Hypoxylon* and allied *Xylariaceae* studied intensively for such compounds in the past decades, there are not many examples for the parallel occurrence of the same compound

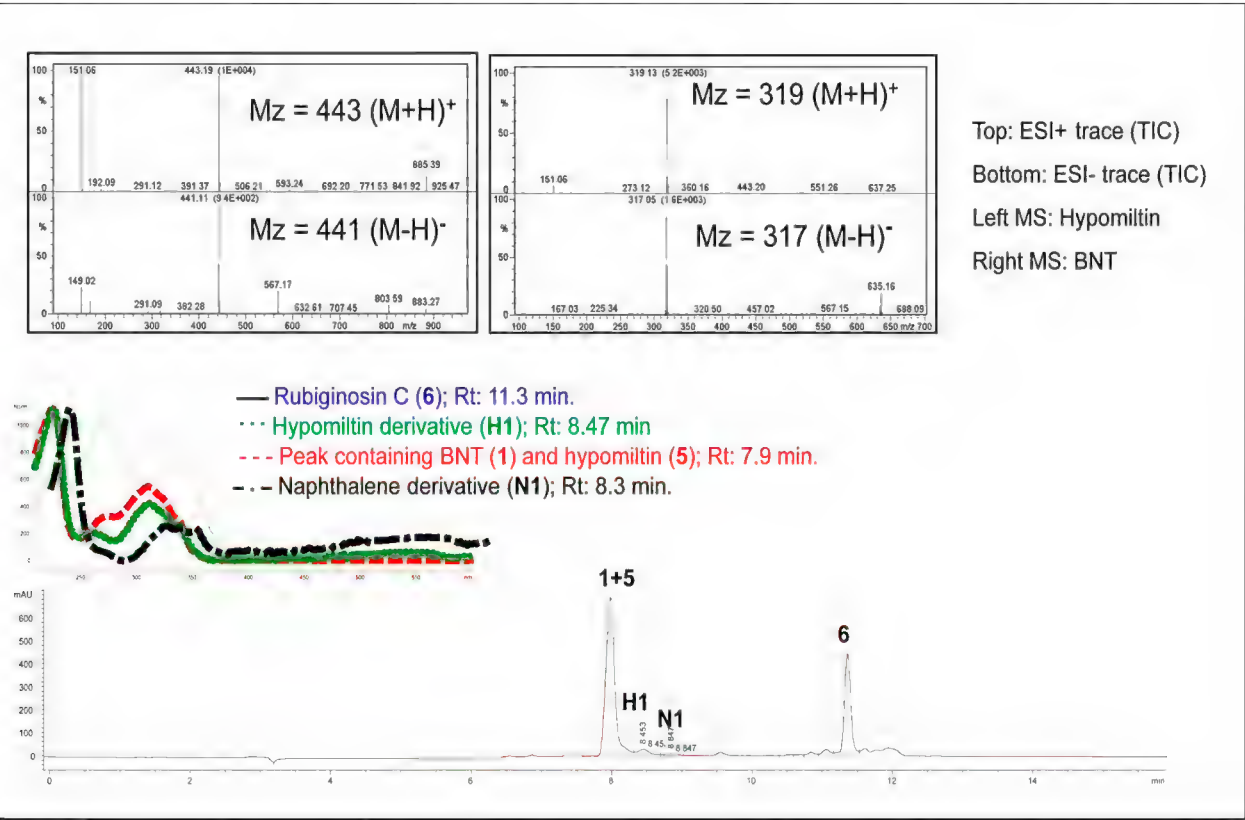


FIG. 13. Stromatal HPLC-UV profile (210 nm) of the stromatal methanol extract derived from the holotype specimen of *Hypoxylon gibriacense*, including DAD and ESI-MS spectra of some major metabolites. Rubiginosin C (6), a peak containing hypomiltin (5) overlaid by BNT (1), and other yet unidentified derivatives of hypomiltin (H1) and BNT (N1) were the major detectable components.

classes in both groups. However, the major stromatal constituents of *H. aeruginosum* J. H. Mill. and other *Xylariaceae* featuring blue or green stromatal surfaces have been recently identified as derivatives of the lichen constituent, lepraric acid (Læssøe et al. 2010).

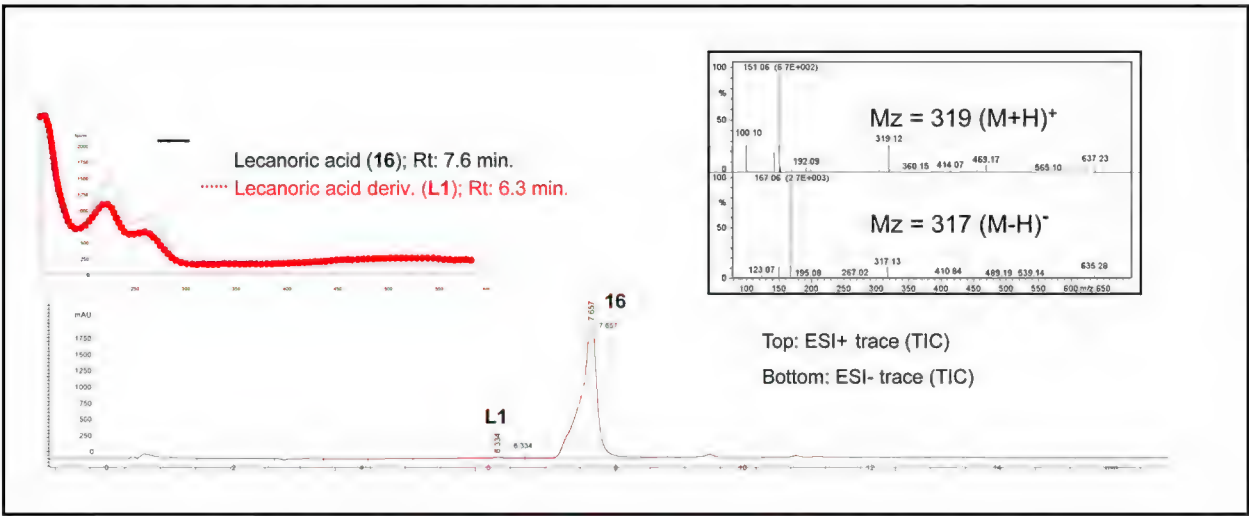


FIG. 14. HPLC-UV profile (210 nm) of the stromatal methanol extract derived from the holotype specimen of *Hypoxylon addis*, including DAD and ESI-MS spectra of some major metabolites. Lecanoric acid (16) was clearly the major detectable component, accompanied by a derivative (L1), but no known metabolites of *Hypoxylon* were detected

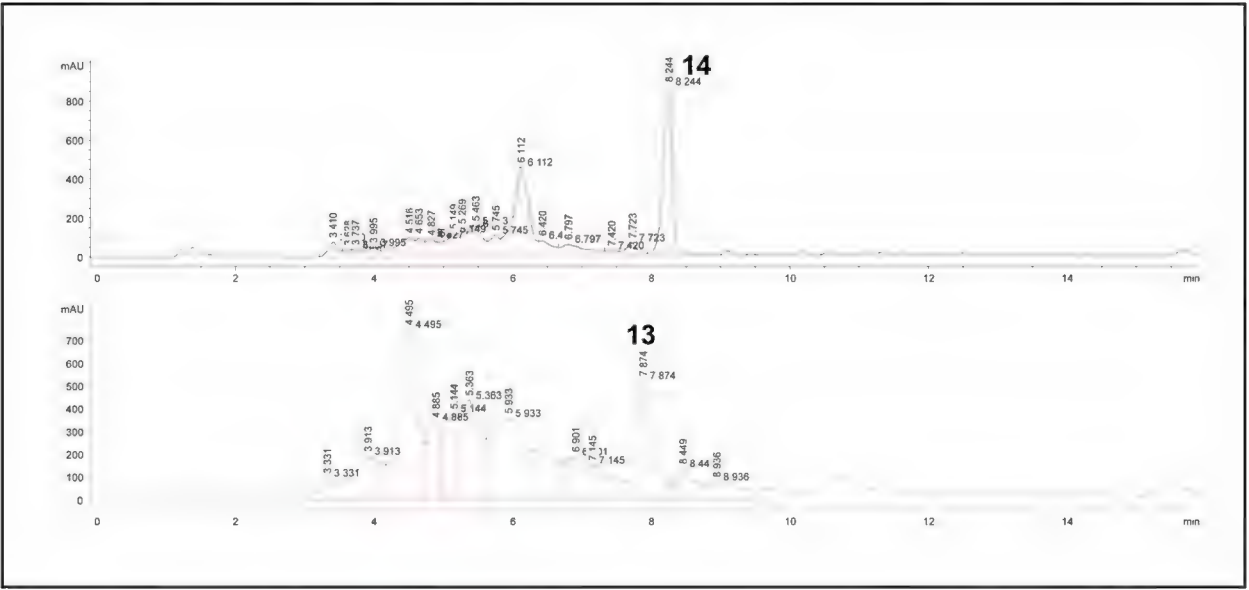


FIG. 15. HPLC-UV profile (210 nm) of the ethyl acetate extracts prepared from YMG cultures of the ex-holotype strains of *Hypoxylon fuscoides* and *H. gibriacense* after 8 days of cultivation, according to Bitzer et al. (2008). The HPLC profile of *H. fuscoides* (above) revealed .5-methylmellein (14) as major component, while *H. gibriacense* (below) produced mellein (13) and a series of other, mostly unknown compounds. The HPLC profile of *H. lusitanicum* (data not shown) closely resembled that of *H. fuscoides* and, therefore, most other members of the *H. fuscum* /*H. rubiginosum* complexes so far studied.

An updated key to European species of *Hypoxylon*

We found it practical to update our key to the species of *Hypoxylon* that have so far been encountered from regions that politically or geographically belong to Europe. This key is based on the one published by Stadler et al. (2004) but taking new results on the chorology of the species into account. In addition, the key incorporates recently published species (Stadler et al. 2008) as well as those newly described in the present study.

Species of *Annulohypoxylon* (formerly regarded as *Hypoxylon* sect. *Annulata* sensu Ju & Rogers 1996), however, have been expelled from the key published in 2004; for morphological characters and differences to *Hypoxylon*, see Hsieh et al. (2005). To safely identify hypoxylloid specimens with papillate ostioles [*A. cohaerens* (Pers.) Y.M. Ju et al. = *H. cohaerens* (Pers.) Fr.; *A. minutellum* (Sydow & P. Sydow) Y.M. Ju et al. = *H. cohaerens* var. *microsporum* J.D. Rogers & Cand.; *A. multiforme* (Fr.) Y.M. Ju et al. = *H. multiforme*(Fr.) Fr.] and with ostioles encircled by a disk [*A. michelianum* (Ces. & De Not.) Y.M. Ju et al. = *H. michelianum* Ces. & De Not.; *A. stygium* var. *annulatum* (Rehm) Y.M. Ju et al. = *H. stygium* var. *annulatum* (Rehm) Y.M. Ju & J.D. Rogers], a comparison with certain *Annulohypoxylon* species keyed by Stadler et al. (2004) as *Hypoxylon* therefore remains indispensable.

- 1 Mature stromata carbonaceous, black, without KOH-extractable pigments [but immature stromata orange, with Dark Purple (80) to Dark Vinaceous (82) pigments]. Ascospores  $9.5\text{--}11.5 \times 4\text{--}5.5 \mu\text{m}$ . (USA, France) ..... *H. submonticulosum* Y.M. Ju & J.D. Rogers
- 1 Mature stromata waxy to woody, not carbonaceous, colored other than black and with KOH-extractable pigments ..... 2
- 2(1) Stromata hemispherical to almost spherical ..... 3
- 2 Stromata effused to pulvinate ..... 6
- 3(2) Stromatal surface Vinaceous Grey (116), Sepia (63) or Grayish Sepia (106), KOH-extractable pigments Pure Yellow (14), Greenish Yellow (16) or Citrine (13), ascospores  $17\text{--}22 \times 9\text{--}11 \mu\text{m}$ ; on *Fraxinus* ..... *H. fraxinophilum* Pouzar
- 3 Stromatal surface Rust (39), Bay (6), or Dark Brick (60), KOH-extractable pigments orange (7), ascospores averaging less than  $15 \mu\text{m}$  long ..... 4
- 4(3) Ascal apical ring present, amyloid; widespread ..... 5
- 4 Ascal apical ring absent; monotypic, not recorded since 1867, ascospores  $9\text{--}11 \times 4.5\text{--}5.5 \mu\text{m}$  ..... *H. commutatum* Nitschke
- 5(4) Mainly on *Fagus*; ascospores  $11\text{--}13.5 \times 5\text{--}6.5 \mu\text{m}$  ..... *H. fragiforme* (Pers.: Fr.) J. Kickx f.
- 5 On other hosts, rarely on *Fagus*; ascospores  $7\text{--}9 \times 3.5\text{--}5 \mu\text{m}$  ..... *H. howeanum* Peck



- 6(2) Stromatal surface with Purple (35) or Vinaceous (57) colors. ....7
- 6 Stromatal surface with Orange (7), Rust (39), Brick (60), or brown colours ... 17
- 6 Stromatal surface with a greenish tone, Isabelline (65), without visible colored granules beneath surface but with Fawn (87) to dilute Umber (9) KOH-extractable pigments; ascospores  $11-12.5(-13.5) \times 6-6.5 \mu\text{m}$ , ellipsoid-equilateral with straight germ slit, perispore indehiscent in 10% KOH (M.S. & J.F.; unpublished data on specimen in K collected in Poland; identified by Z. Pouzar). .... *H. papillatum* Ellis & Everh.
- 7(6) Ascospores averaging more than  $20 \mu\text{m}$  long. .... 8
- 7 Ascospores averaging less than  $15 \mu\text{m}$  long ..... 9
- 8(7) KOH-extractable pigments Pale Vinaceous Grey (115) to Vinaceous Grey (116) in fresh specimens or absent in aged specimens; ascospores  $18.5-23 \times 8-10 \mu\text{m}$  with germ slit spore-length. .... *H. vogesiacum* (Pers.) Sacc.
- 8 Boreal distribution, frequently on *Salix*, KOH-extractable pigments dense, Greenish Olivaceous (90); ascospores  $22-31 \times 8.5-11 \mu\text{m}$  with faint germ slit less than spore-length ..... *H. macrosporum* P. Karst.
- 9(7) KOH-extractable pigments Livid Purple (81) or absent ..... 10
- 9 KOH-extractable pigments pigments Orange (7) to Sienna (8). .... 11
- 9 KOH-extractable pigments Amber (47), Isabelline (65), Olivaceous (48), Gray Olivaceous (107), Greenish Olivaceous (90), Citrine (13) or otherwise with yellow, green or brown tones. .... 13
- 10(9) Stromata effused,  $0.4-0.5 \text{ mm}$  thick, with KOH-extractable pigments dilute, Livid Purple (81) or absent; ascospores  $8-11.5 \times 4.5-5 \mu\text{m}$  with a straight germ slit. Distribution world-wide; major stromatal metabolites: carneic acids (Quang et al. 2006) ..... *H. carneum* Petch
- 10 Stromata pulvinate,  $0.8-1.4 \text{ mm}$  thick, with KOH-extractable pigments Vinaceous Purple (101); ascospores  $9.5-12.5 \times 5-6 \mu\text{m}$  with a sigmoid germ slit (France, UK, present study) ..... *H. fuscoide*s
- 11(9) Ascospores  $11-13.5 \times 5-7 \mu\text{m}$ , with perispore faintly striate by LM; known from Portugal (present study) ..... *H. lusitanicum*
- 11 Ascospores  $9.5-11.5 \times 4.5-6 \mu\text{m}$ , with perispore smooth by LM ..... 12
- 12(11) Stromata widely effused with jagged black margins; ascospores  $9-11.5 \times 5-6 \mu\text{m}$ ; host preference for *Fraxinus*; Temperate Europe and USA (Stadler et al. 2008) . .... *H. petriniae* M. Stadler & J. Fourn.
- 12 Stromata less widely effused to effused-pulvinate, with concolorous margins; ascospores  $9.5-11.5 \times 4.5-5 \mu\text{m}$ ; known from the Canary Islands (Stadler et al. 2008) ..... *H. canariense* J. Fourn. et al.
- 13(9) Perithecia obovoid to frequently tubular, up to  $1 \text{ mm}$  high; stromatal surface with a metallic shine when mature. Recorded from Central and Western Europe and North America (various hosts). Ascospores  $9.5-11.5 \times 4-4.8 \mu\text{m}$  ..... *H. macrocarpum* Pouzar
- 13 Perithecia spherical to obovoid, not tubular; stromatal surface lacking a metallic shine when mature ..... 14

- 14(13) Ascospores with straight germ slit. . . . . 15
- 14 Ascospores with slightly sigmoid germ slit. . . . . 16
- 15(14) Ascospores ellipsoid-inequilateral in lateral view,  $9\text{--}12 \times 4\text{--}6\ \mu\text{m}$ , perispore dehiscent in 10% KOH. KOH-extractable pigments Amber (47), Greenish Yellow (16) or Citrine (13) . . . . . *H. perforatum* (Schwein.) Fr.
- 15 Ascospores ellipsoid, nearly equilateral in lateral view, often pyriform,  $12\text{--}15 \times 5.5\text{--}7\ \mu\text{m}$ , perispore indehiscent in 10% KOH. KOH-extractable pigments Olivaceous (48), Greenish Olivaceous (90), Gray olivaceous (127), or Olivaceous Gray (121). So far known from Austria, Germany, Slovakia, and North America. . . . . *H. fuscopurpureum* (Schwein.) M.A. Curtis
- 16(14) Stromata with pure yellow (14) to luteous (12) granules and greenish olivaceous (90) KOH-extractable pigments; apparently restricted to *Quercus*, with a boreal distribution; ascospores  $10\text{--}13.5 \times 4\text{--}5\ \mu\text{m}$ . So far known from France (Stadler et al. 2004), Scandinavia (Granmo 1999) and USA (Stadler et al. 2008) . . . . . *H. porphyreum* Granmo
- 16 Stromata with sienna (8) or otherwise orange brown granules and KOH-extractable pigments amber (47), isabelline (65), olivaceous (48), gray olivaceous (107), or greenish olivaceous (90); widespread, preferably on *Betulaceae* and other hosts, but not yet safely recorded from *Quercus*; ascospores  $11\text{--}16 \times 5\text{--}8\ \mu\text{m}$  . . . . . *H. fuscum* (Pers.) Fr.
- 17(6) Young stromata with a bright yellow to orange fimbriate margin; perithecia small,  $0.1\text{--}0.3\ \text{mm}$  diam, seated on a well developed black basal tissue . . . . . 18
- 17 Young stromata lacking a bright yellow to orange fimbriate margin. . . . . 19
- 18(17) Ascospores  $5\text{--}7 \times 2.5\text{--}3.5\ \mu\text{m}$ , ellipsoid-inequilateral in lateral view. So far recorded from Austria, Croatia, Germany (Bitzer et al. 2008). France, Italy, Slovakia (Ripková & Hagara 2003) and Switzerland . . . . . *H. ticinense* L.E. Petrini
- 18 Ascospores  $8\text{--}11 \times 4\text{--}5\ \mu\text{m}$ , ellipsoid-equilateral in lateral view. So far recorded from France and USA. . . . . *H. subticinense* Y.M. Ju & J.D. Rogers
- 19(17) KOH-extractable pigments yellow or orange. . . . . 20
- 19 KOH-extractable pigments with shades of olivaceous brown . . . . . 33
- 20(19) Ascal apical ring highly reduced or lacking, inamyloid . . . . . 21
- 20 Ascal apical ring present, amyloid. . . . . 23
- 21(20) Stromata glomerate with ostioles encircled by a ring; ascospores  $11.5\text{--}13 \times 6\text{--}6.8\ \mu\text{m}$  with perispore conspicuously striate by LM (France, present study) . . . . . *H. gibriacense*
- 21 Stromata applanate to pulvinate, with ostioles lacking a ring; ascospores with perispore smooth by LM . . . . . 22
- 22(21) Stromata discoid, encircled with a swollen stellate margin, on bark of *Fraxinus*; ascospores  $9.5\text{--}12 \times 5\text{--}6\ \mu\text{m}$ . Europe and North America . . . . . *H. cercidicola* (Berk. & M.A. Curtis ex Peck) Y.M. Ju & J.D. Rogers
- 22 Stromata pulvinate to hemispherical, reported from *Carpinus*, ascospores  $9\text{--}11 \times 4.5\text{--}5.5\ \mu\text{m}$  (see also 4). . . . . *H. commutatum* Nitschke

23(20)	Ascospores averaging more than 14 µm long . . . . .	24
23	Ascospores averaging less than 12 µm long . . . . .	25
24(23)	Stromata pulvinate restricted at base, with rust brown to ochraceous brown granules beneath surface; apparently rare, known only from <i>Tilia</i> and <i>Sorbus</i> ; ascospores 14–17 × 6.5–8 µm, Recorded from Switzerland (Petrini & Müller 1986), Slovakia and Canada (Stadler et al. 2008). . . . .	<i>H. ferrugineum</i> G. H. Otth
24	Stromata effused to pulvinate, with blood red granules beneath surface, recorded from various hosts; ascospores 15–18 × 6–7.5 µm . . . . .	<i>H. julianii</i> L.E. Petrini
25(23)	KOH-extractable pigments Amber (47), Greenish Yellow (16) or Citrine (13); ascospores 9.7–11.5 × 4.7–5.3 µm (see also 15) . . . . .	<i>H. perforatum</i> (Schwein.) Fr.
25	KOH-extractable pigments Orange (7), Sienna (8), Rust (39) or Scarlet (5) . . .	26
26(25)	Ascospores almost equilateral in lateral view. . . . .	27
26	Ascospores inequilateral in lateral view . . . . .	28
27(26)	Ascospores 7–10 × 3–4.5 µm; stromatal surface dark rust (39) to sepia (63), with dark orange granules beneath surface and KOH-extractable pigments fulvous (43) to rust (39); recorded from <i>Salix</i> in Northern Europe (Granmo 1999), Belgium (J.F. & M.S., unpublished data), and USA (Ju & Rogers 1996, as unnamed segregate in “Notes to <i>H. rubiginosum</i> ”) . . . . .	<i>H. salicicola</i> Granmo
27	Ascospores 9.5–12.5 × 4.8–6 µm; stromatal surface vinaceous buff (86), greyish sepia (106) to brown vinaceous (84) with bright yellow granules beneath surface and between perithecia; KOH-extractable pigments hazel (88), sienna (8) to umber (9). Apparently restricted to <i>Sorbus</i> , with a boreal distribution (Granmo 2001) . . . . .	<i>H. liviae</i> Granmo
28(26)	Ascospores averaging less than 11 µm long . . . . .	29
28	Ascospores averaging more than 11 µm long. . . . .	31
29(28)	Stromata with papillate ostioles. . . . .	30
29	Stromata with umbilicate ostioles . . . . .	<i>H. rubiginosum</i> (Pers.) Fr.
30(29)	Stromata erumpent, pulvinate, small, with orange granules beneath surface; known from Europe and USA, restricted to <i>Populus</i> ; ascospores 8–10 × 3.5–4.5 µm . . . . .	<i>H. laschii</i> Nitschke
30	Stromata superficial, effused to pulvinate, with blood red granules beneath surface; distribution apparently world-wide, without apparent host specificity ascospores 7.5–10 × 4–4.8 µm. . . . .	<i>H. rutilum</i> Tul. & C. Tul.
31(28)	Perithecia up to 0.2 mm diam; ascospores 11–14.5 × 5–6 µm with slightly sigmoid germ slit; Canary Islands . . . . .	<i>H. urriesii</i> J. Fourn. & M. Stadler
31	Perithecia 0.3–0.45 mm diam; ascospores with straight germ slit . . . . .	32
32(31)	Ascospores 9.5–11.5 × 4.5–5 µm; known from the Canary Islands (see also 12) . . . . .	<i>H. canariense</i> J. Fourn. et al.
32	Ascospores 11–13.5 × 5–7 µm; known from Portugal (see also 11) . . . . .	<i>H. lusitanicum</i>

- 33(18) KOH-extractable pigments Hazel (88), Sienna (8) to Umber (9);  
perithecia obovoid, up to 0.4 mm high; ascospores dark brown, ellipsoid,  
nearly equilateral,  $9.5\text{--}12.5 \times 4.8\text{--}6\ \mu\text{m}$ ; apparently restricted to *Sorbus*  
(see also 27) ..... *H. liviae* Granmo
- 33 KOH-extractable pigments isabelline (65), umber (9), or grayish sepia (106);  
perithecia frequently tubular, up to 1 mm high; ascospores inequilateral and  
narrower,  $9.5\text{--}11.5 \times 4\text{--}4.8\ \mu\text{m}$  (see also 13). ..... *H. macrocarpum* Pouzar

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## New records of graminicolous smut fungi in Ukraine

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**Abstract** – Four new records of graminicolous smut fungi are reported from Ukraine. Among these species *Tranzscheliella hypodytes* and *Urocystis agrostidis* were found on new host plants, *Ustilago aeluropodis* and *Ustilago trebouxii* are new fungi for Ukraine.

**Key words** – *Ustilaginomycetes*, *Poaceae*

### Introduction

The smut fungi of Ukraine have not been investigated exhaustively. Studies on these fungi have been carried out intensively in only two regions— Halitzcia, by Polish mycologists (Wróblewski 1912, 1915, 1922; Raciborski 1910, Krupa 1888), and around Kyiv (Zelle 1925, Lavitska 1949, 1978). Other regions of Ukraine are poorly investigated for smut fungi. Southern Ukraine is part of the steppe zone. The true steppe is a unique natural phenomenon that is preserved only in Ukraine and Russia. The protected remnants of the Ukrainian steppe have a high level of endemism in local plant communities. Therefore, this region of the country is promising in terms of discovery of new species of plant pathogenic micromycetes, including smut fungi. Another interesting region of Ukraine is Volhynian Polissia situated in the northwest part of the country. Ancient forests and peat bogs contribute to great floristic diversity in this area. Thus, it is not surprising that the first detailed examination of these fungi in Ukraine provided noteworthy results. In this paper, we examined a collection from Volhynian Polissia containing two specimens of a smut fungus infecting *Melica ciliata* L. and specimens of smut fungi from the steppe region in the southern part of the country.

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\* corresponding author

## Materials and methods

Sori, spore balls, and spores were studied using dried herbarium specimens. For light microscopy (LM), spore balls and spores were dispersed in a droplet of lactophenol on a microscope slide, covered with a cover glass, gently heated to boiling point to rehydrate the spores and eliminate air bubbles, and examined at 400× and 1000× magnification. For scanning electron microscopy (SEM), spores were placed on double-sided adhesive tape, mounted on a specimen stub, sputter-coated with gold, ca. 15 nm, and examined in SEM at 30 kV.

## Results and discussion

Only 45 species of smut fungi on plants belonging to different grass genera have been reported in Ukraine. The genus *Ustilago* (Pers.) Roussel is represented by 18 species, *Sporisorium* Ehrenb. ex Link by 7 species, the genera *Tilletia* Tul. & C. Tul. and *Urocystis* Rabenh. ex. Fuckel by 6 species each, *Tranzscheliella* Lavrov by 3 species, and *Jamesdicksonia* Thirum. et al., *Macalpinomyces* Langdon & Full., *Moesziomyces* Vánky, *Neovossia* Körn., and *Ustilentyloma* Savile by one species each (Zerova et al. 1971, Savchenko et al. in press). In adjacent countries, for example Poland, 44 species of graminicolous smut fungi were reported (Piątek et al. 2005), and more than 70 species of graminicolous smut fungi were reported in Russia (Karatygin & Azbukina 1989, Azbukina et al. 1995). As a result of our studies on recently collected specimens in the Volhynian Polissia and southern steppe regions of Ukraine we have extended the list of Ukrainian graminicolous smut fungi to include *Urocystis agrostidis*, *Ustilago aeluropodis*, and *U. trebouxii*. Another species, *Tranzscheliella hypodytes*, is reported on a new host plant.

### *Tranzscheliella hypodytes* (Schltld.) Vánky & McKenzie

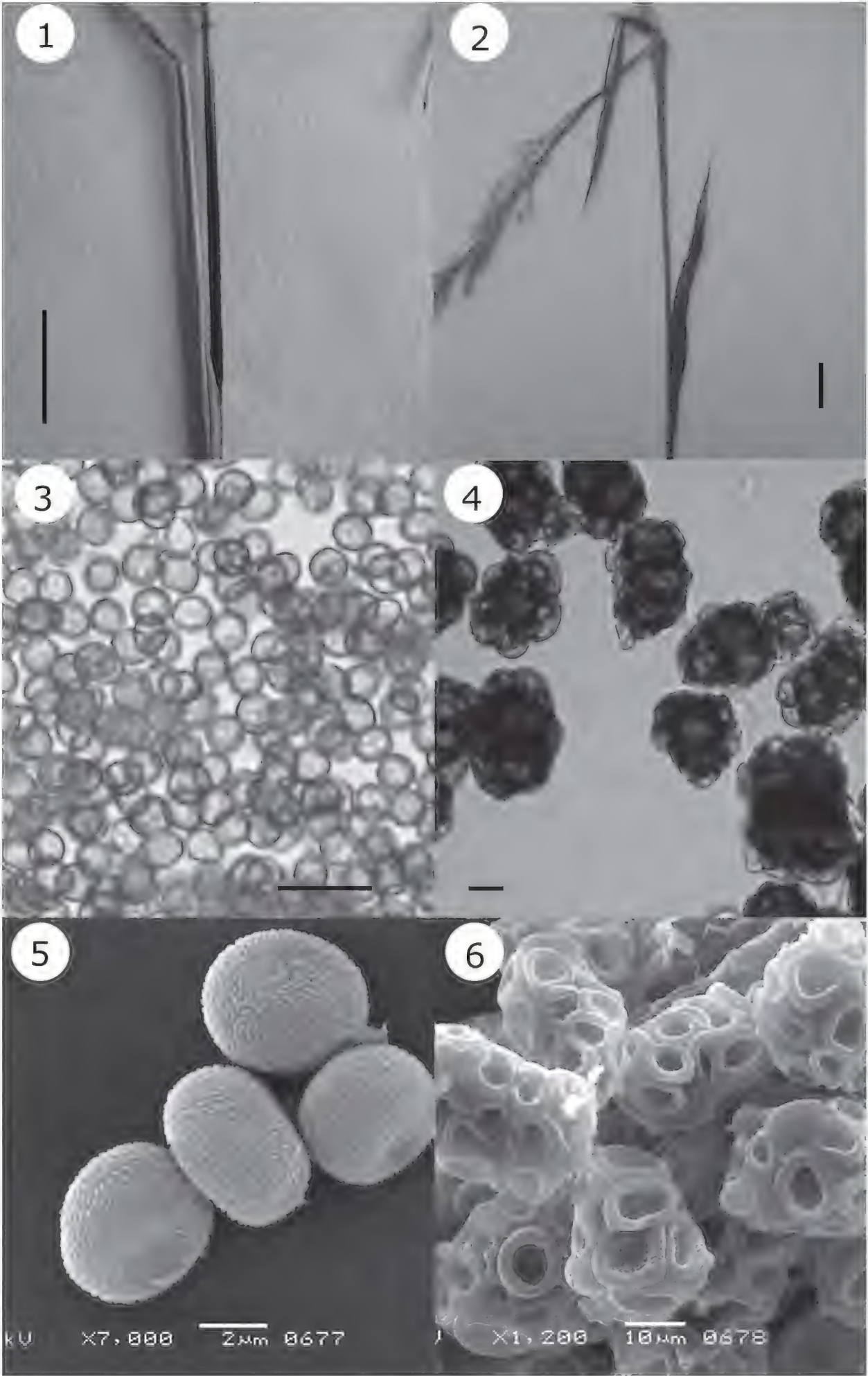
FIGS. 1, 3, 5

SORI in culms as a black to blackish brown, semi-agglutinated to powdery spore mass surrounding the upper internodes and in the axis of abortive inflorescences. Sori first hidden by a leaf-sheath, then naked. Infection systemic, inflorescences usually sterile. SPORES globose, subglobose to elongated, irregular or slightly to medium compressed,  $3.9\text{--}5.2 \times 4.6\text{--}5.7 \mu\text{m}$  (at the mean  $4.6 \times 5.2 \mu\text{m}$ ), medium to dark olivaceous brown. Spore wall smooth, ca.  $0.5 \mu\text{m}$  thick, in SEM densely and minutely verruculose on the wall surface, hyaline cap occasionally present.

SPECIMEN EXAMINED — On *Elymus uralensis* subsp. *viridiglumis* (Poaceae): Ukraine, Kherson region, Hola Prystan district, Black Sea Biosphere Reserve, 6.VII.2009, K. & M. Savchenko, KW 36370F.

FIGS. 1–6. 1, 3, 5: *Tranzscheliella hypodytes* on *Elymus uralensis* subsp. *viridiglumis*; 1: part of infected plant with sori; 3, 5: spores in LM and SEM. 2, 4, 6: *Urocystis agrostidis* on *Agrostis gigantea* subsp. *maeotica*; 2: infected plant with sori; 4, 6: spore balls in LM and SEM. Bars: 1, 2 = 1 cm; 3, 4, 6 = 10  $\mu\text{m}$ ; 5 = 2  $\mu\text{m}$ .





COMMENTS: *Tranzscheliella hypodytes* was collected on a new host plant, *Elymus uralensis* subsp. *viridiglumis* (Nevski) Tzvelev (= *Agropyron lavrenkoanum* Prokudin). This fungus has an extremely broad host range that includes a number of *Elymus* spp., but this is the first report on *E. uralensis*.

***Urocystis agrostidis* (Lavrov) Zundel**

FIGS. 2, 4, 6

SORI in leaves as long streaks between the veins, initially lead-colored and covered by the epidermis of the host plant which later ruptures longitudinally and the black powdery mass of spore balls becomes scattered. SPORE BALLS globose, subglobose,  $22\text{--}50 \times 25\text{--}50\ \mu\text{m}$  in diameter, composed of 1–4 central spores and a continuous layer of sterile cells. SPORES globose to subglobose,  $13\text{--}17 \times 15\text{--}20\ \mu\text{m}$ , olivaceous brown to light brown with a smooth surface. Sterile cells ovoid, subglobose to elongated,  $7\text{--}15\ \mu\text{m}$  long, pale yellowish to reddish brown.

SPECIMEN EXAMINED — On *Agrostis gigantea* subsp. *maeotica* (Poaceae): Ukraine, Kherson region, Hola Prystan district, Black Sea Biosphere Reserve, 4.VII.2009, K. & M. Savchenko, KW 36369F.

COMMENTS: During a trip to the Black Sea Biosphere Reserve (steppe zone, South of Ukraine) in 2009 a smutted specimen of *Agrostis gigantea* subsp. *maeotica* (Klokov) Tzvelev was collected. The fungus was identified as *Urocystis agrostidis*. Until now, no *Urocystis* species on *Agrostis* L. have been reported in Ukraine. This represents a new species of smut fungus for Ukraine and a new host record for the species.

***Ustilago aeluropodis* (Trotter) Vánky**

SORI on the tip of culms replacing the inflorescences, 1–2 cm long, hidden by leaf-sheaths and young leaves, with maturation rupturing to expose the dark brown, powdery spore mass. Peridium, sterile cells, and columellae are lacking. SPORES very variable in shape and size, in certain cases aggregated into loose, ephemeral spore groups, globose, ellipsoidal,  $10\text{--}17 \times 12\text{--}18\ \mu\text{m}$ , golden brown to brown, wall  $0.8\text{--}1.5\ \mu\text{m}$  thick, in LM sparsely to moderately densely, minutely verruculose; in SEM minutely verruculose-echinulate.

SPECIMEN EXAMINED — On *Aeluropus littoralis* (Poaceae): Ukraine, Kherson region, Hola Prystan district, Black Sea Biosphere Reserve, 18.VIII.2003, O.Yu. Umanets, KW 36361F.

COMMENTS: During the last ten years phytotrophic micromycetes have been collected by Dr. O. Umanets in the Black Sea Biosphere Reserve (Kherson region, Ukraine). Among the specimens examined, we found several examples of vascular plants infected by smut fungi. Previously we reported *Sporisorium cenchri* (Lagerh.) Vánky as a new record for Ukraine (Savchenko et al., in press). In 2003 Dr. O. Umanets also collected infected plants of *Aeluropus littoralis*

(Gouan) Parl. We have identified the smut as *Ustilago aeluropodis*, another new record for Ukraine. Prior to this record, this species was reported in Europe only from Romania (Vánky 1994).

***Ustilago trebouxii* Syd. & P. Syd.**

SORI in the upper leaves and leaf-sheaths as long, dark to olivaceous brown striae. Spore mass initially covered by the epidermis, exposed with maturation, becoming powdery. SPORES subglobose, rarely globose to slightly irregular,  $3.5\text{--}5 \times 4\text{--}6 \mu\text{m}$ , pale olivaceous brown, in LM smooth, in SEM sparsely minutely punctuate-verruculose; verrucae never merged.

SPECIMENS EXAMINED — On *Melica ciliata* (*Poaceae*): Ukraine, Volhynian region, Ratne district, near Sviate lake, 1.VIII.2009, K. Savchenko, KW 36854F, 36855F.

COMMENTS: It is not inconceivable that this fungus will be also found in Poland and Byelorussia.

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We are grateful to Dr. Kálmán Vánky (Herbarium *Ustilaginales* Vánky, Tübingen, Germany) and Dr. Lori Carris (Department of Plant Pathology, Washington State University, Pullman, USA) for reading the manuscript and serving as pre-submission reviewers, to Dr. Lori Carris also for checking our English and helpful comments, to Dr. Olga Umanets for the kindly sent specimen of smutted plant and support during the author's trip to Black Sea Biosphere Reserve, and to Mr. Viktor Novychenko for assistance with the SEM photographs.

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***Bionectria vesiculosa* sp. nov. from Yunnan, China**

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**Abstract** — A new species of *Bionectria* on decaying leaves is described from Xishuangbanna in southwestern China. It is distinctive in the brown perithecia, laterally collapsing when dry, a ring of large vesicular cells surrounding the subapical region of the perithecia, clavate asci with an apical ring, and fusiform, smooth-walled ascospores. Morphology and sequence analysis of ITS and 28S nrDNA support its taxonomic position as a new species in *Bionectria*.

**Key words** — Chelex-100, taxonomy

**Introduction**

The genus *Bionectria* Speg. (*Bionectriaceae*) is characterized by pale yellowish to orange perithecia that do not change color in 3% KOH or lactic acid, a smooth to warted perithecial wall of 1–3 layers, clavate asci with or without an apical ring, 2-celled ascospores with smooth, spinulose, warted or striate surface, and *Clonostachys* anamorphs. Members of *Bionectria* occur on woody and herbaceous plants or other fungi, and are mainly distributed in tropical and subtropical regions (Rossman et al. 1999, Schroers 2001). In connection with our work on the Chinese fungus flora, an interesting fungus was encountered that has brown perithecia with a ring of large cells at the subapical region. On the basis of the teleomorph morphology and sequence analysis of two nuclear ribosomal genes (nrDNA), its position in *Bionectria* is confirmed; its relationship with other species of the genus is discussed.

**Materials & methods**

The taxonomic treatments and methods of Rossman et al. (1999) and Schroers (2001) were followed for the morphological study. Water was used as the mounting medium

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\* Correspondence author

TABLE 1. Sequences analyzed to determine relationships among species in *Bionectria*.

SPECIES	ITS nrDNA		28S nrDNA	
	Collection no.	GenBank no.	Collection no.	GenBank no.
<i>Bionectria compactiuscula</i> Schroers	CBS 913.97	AF358245	CBS 919.97	AF210690
<i>B. coronata</i> (Juel) Schroers	CBS 696.93	AF210667	CBS 696.93	AF210667
<i>B. epichloe</i> (Speg.) Schroers	CBS 101037	AF210675	CBS 101037	AF210675
<i>B. grammicospora</i> (Ferd. & Winge) Schroers & Samuels	CBS 209.93	AF210678	CBS 209.93	AF210678
<i>B. grammicosporopsis</i> (Samuels) Schroers & Samuels	CBS 115.67	AF210679	CBS 115.67	AF210679
<i>B. levigata</i> Schroers	CBS 948.97	AF210680	CBS 948.97	AF210680
<i>B. ochroleuca</i> (Schwein.) Schroers & Samuels	CBS 194.57	AF358249	CCFC 226708	AY686634
<i>B. pityrodes</i> (Mont.) Schroers	CBS 246.78	AF210673	CBS 102033	AF210672
<i>B. ralfsii</i> (Berk. & Broome) Schroers & Samuels	CBS 102845	AF358253	CBS 129.87	AF210676
<i>B. rossmaniae</i> Schroers	CBS 210.93	AF358227	CBS 211.93	AF210665
<i>B. sesquicillii</i> (Samuels) Schroers	CBS 180.88	AF210666	CBS 180.88	AF210666
<i>B. setosa</i> Schroers	CBS 834.91	AF210670	CBS 834.91	AF210670
<i>B. vesiculosa</i>	HMAS 183151	<b>HM050304<sup>a</sup></b>	HMAS 183151	<b>HM050302</b>
<i>B. zelandiae-novae</i> Schroers	CBS 100979	AF358229	CBS 232.80	AF210684
<i>Clonostachys divergens</i> Schroers	CBS 967.73b	AF210677	CBS 967.73b	AF210677
<i>C. miodochialis</i> Schroers	CBS 997.69	AF210674	CBS 997.69	AF210674
<i>C. phyllophila</i> Schroers	CBS 685.96	AF210663	CBS 921.97	AF210664
<i>Hydropisphaera erubescens</i> (Roberge ex Desm.) Rossman & Samuels	HMAS 91779	FJ969800	HMAS 91779	GU075862
<i>Ijuhya paraparilis</i> (Samuels) Rossman & Samuels	HMAS 183506	FJ969801	HMAS 183506	<b>HM050303</b>

<sup>a</sup> Numbers in bold indicate the newly submitted sequences.

for microscopic examinations and measurements; and photographs were taken from water or cotton blue mounts (Stevens 1981). Continuous measurements of individual structures are based on 30 units except as otherwise noted. Specimen examined is deposited in the Mycological Herbarium, Institute of Microbiology, Chinese Academy of Sciences (HMAS).

Chelex-100 was applied to extract genomic DNA from the dehydrated perithecia according to the method by Zhang et al. (2006) with modifications. Fifty perithecia were carefully collected from the substrate with a pair of forceps and rinsed in sterilized water. The perithecia were transferred into a 1.5 ml eppendorf tube, mixed with equal volume of quartz sand, and thoroughly ground with a glass pestle for 10 min. Then 200 µl 10%

w/v Chelex-100 chelating resin (Sigma) was added and mixed for 10 sec on a vortex. The tube was incubated at 56°C for 2 hr, mixed for 10 sec, and then incubated at 99°C for 10 min. After centrifuging at 12000 r/min for 10 min, the supernatant was transferred to another 1.5 ml tube filled with 4/5 volume of 100% pre-cooling isopropanol. The mixture was placed at –20°C overnight, and centrifuged at 12000 r/min for 15 min. After rinsing with 200 µl 75% ethanol, the precipitant was dried at room temperature and dissolved in 30 µl TE or ddH<sub>2</sub>O as PCR template.

The ITS1-5.8S-ITS2 (ITS) and 28S regions of the nrDNA were amplified by using the primer pairs, ITS5–ITS4 (White et al. 1990), and LROR–LR5 (Rehner & Samuels 1994, Vilgalys & Hester 1990). The PCR reaction mixture (50 µl) contained 5.0 µl 10× PCR buffer, 3.0 µl MgCl<sub>2</sub> (25 mM), 2.5 µl sense primer (10 µM), 2.5 µl antisense primer (10 µM), 1.0 µl dNTP (10 mM each), 2.5 µl DNA template, 0.5 µl Taq polymerase (5.0 U/µl) (Bio Basic Inc.) and 33 µl ddH<sub>2</sub>O. Reactions were performed on the 2720 Thermal Cycler (Applied Biosystems) with cycling conditions of denaturation at 95°C for 5 min, followed by 35 cycles of denaturation at 94°C for 30 s, annealing at 52°C (ITS region) and at 55°C (28S region) for 30 s and elongation at 72°C for 60 s, with a final extension step at 72°C for 5 min to complete the reactions. Amplicon was purified with the PCR Product Purification Kit (Biocolor BioScience & Technology Co.) and sequenced with the ABI BigDye Terminator V3.1 Cycle Sequencing Kit (Applied Biosystems) on an ABI 3730XL DNA Sequencer (SinoGenoMax Co. Ltd). The amplifying primers were served as sequencing primers. Final sequences were checked and edited manually by using BioEdit V.7.0.5 (Hall 1999). Sequences of the related species were retrieved from GenBank. Materials studied are listed in TABLE 1.

All sequences were aligned using ClustalX V.1.8 (Thompson et al. 1997), and the alignments were visually adjusted while necessary. A Neighbor-Joining tree was generated using MEGA 4.10 (Tamura et al. 2007) based on combined sequences of ITS and 28S genes with *Hydropisphaera erubescens* and *Ijuhya paraparilis* as outgroup taxa. Kimura 2-parameter was selected as the nucleotide substitution model, and gaps or missing data were pairwise deleted. Bootstrap method was performed with 1000 replicates to test phylogeny branch support.

## Results and discussion

*Bionectria vesiculosa* J. Luo & W.Y. Zhuang, sp. nov.

FIGS. 1–2

MYCOBANK MB518120

*Peritheciis subglobosis*, 100–155 µm diam; *ascis clavatis*, 8-sporis, 35–47 × 3.5–7 µm; *ascosporis fusiformibus*, *uniseptatis*, 9.5–13 × 1.5–3 µm.

**HOLOTYPE:** China, Yunnan, Xishuangbanna, on decaying leaves of a dicotyledonous plant, W.-P. Wu & Y. Huang W2728b, 16 X 1999, HMAS 183151.

**ETYMOLOGY:** The specific epithet refers to the vesicular cells forming a ring on the perithecial apex.

Ascomata on white subiculum, perithecial, solitary or gregarious up to 3 in a group, superficial, subglobose, 110–160 µm high, 100–155 µm diam., laterally collapsing when dry, pale yellow when young, and reddish brown to brown at

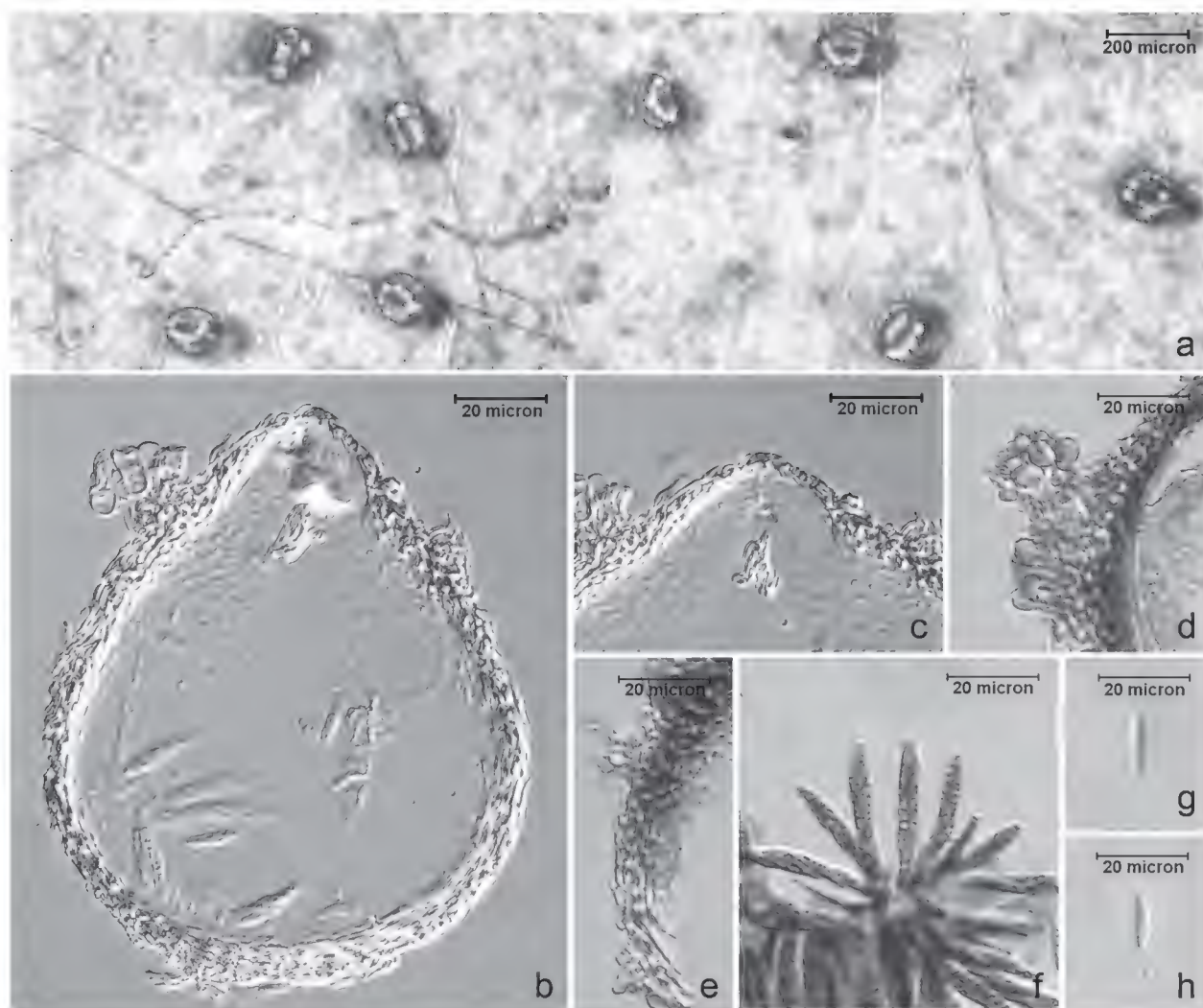


FIG. 1. *Bionectria vesiculosa* (HMAS 183151). a. Ascomata on natural substrate; b. Median section of an ascoma; c. Median section through apical portion of an ascoma; d, e. Structure of ascomatal wall at subapical portion showing vesicular cells; f. Asci with an apical ring; g, h. Ascospore.

maturity, not changing color in 3% KOH or lactic acid, surface smooth, with a ring composed of large cells or a ring of wart-like structures surrounding subapical region; coronate ring 5–26  $\mu\text{m}$  high, pale yellow, cells vesicular, 2–12  $\times$  2–7  $\mu\text{m}$ , cell walls 0.5–1  $\mu\text{m}$  thick. Ascomatal wall 7–15  $\mu\text{m}$  thick, of two layers; outer layer 5–10  $\mu\text{m}$  thick, cells angular, 5–10.5  $\times$  3–5.5  $\mu\text{m}$ , cell walls 0.5–2  $\mu\text{m}$  thick; inner layer 2–6  $\mu\text{m}$  thick, cells flattened, 6–11.5  $\times$  1–3  $\mu\text{m}$ , cell walls 0.5–1.5  $\mu\text{m}$  thick. Asci clavate, 8-spored, with an apical ring, 35–47  $\times$  3.5–7  $\mu\text{m}$  ( $n = 50$ ). Ascospores fusiform, uniseptate, not constricted at septum, hyaline, smooth, with 6–9 guttules, biseriate, 9.5–13  $\times$  1.5–3  $\mu\text{m}$  ( $n = 50$ ).

ANAMORPH: Unknown.

NOTES: Morphologically, the perithecial anatomy and negative reactions to KOH and lactic acid of the new species indicate its position in *Bionectria*. Unlike any other species of the genus, a crown-like ring composed of large vesicular cells is present at the subapical region of the perithecia. *Bionectria vesiculosa* is somewhat similar to *B. setosa* in having brown perithecia less than 200  $\mu\text{m}$  in



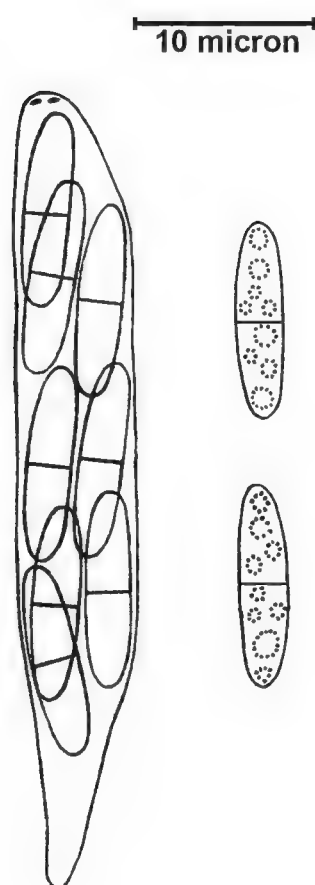


FIG. 2. Ascus and ascospores of *Bionectria vesiculosa* (HMAS 183151).

diam., two-layered perithecial wall, clavate asci with an apical ring, shape, size, septation and surface morphology of ascospores, and leaf-inhabiting. The latter differs in having smooth and thicker perithecial walls 20–30  $\mu\text{m}$  thick and asci 45–53  $\times$  6.5–9  $\mu\text{m}$  (Schroers 2001). The new species also resembles *B. coronata* in the presence of a thin subiculum at the perithecial base, small, subglobose perithecia that are laterally pinched when dry, acute perithecial apex, shape and size of asci, shape, size and surface of ascospores, and foliicolous habit. *Bionectria coronata* differs in having pale yellow to yellowish orange perithecia, one-layered perithecial walls, 15–20  $\mu\text{m}$  thick, with the outermost cell layer connected with a hyphal stroma, undulate setae surrounding the ostiole, asci lacking of an apical apparatus, and unicellular ascospores (Schroers 2001).

Seventeen related species of *Bionectria/Clonostachys* were selected to investigate the phylogenetic position of *B. vesiculosa*. As shown in FIG. 3, all species tested formed one monophyletic clade with 100% bootstrap support, which confirms the placement of the new species in *Bionectria*. The morphologically similar *B. coronata* appears to be only distantly related. *Bionectria pityrodes* and *B. setosa* form a poorly supported subclade with *B. vesiculosa* (FIG. 3). The morphological characteristics of these three species do not show much similarity (Schroers 2001).

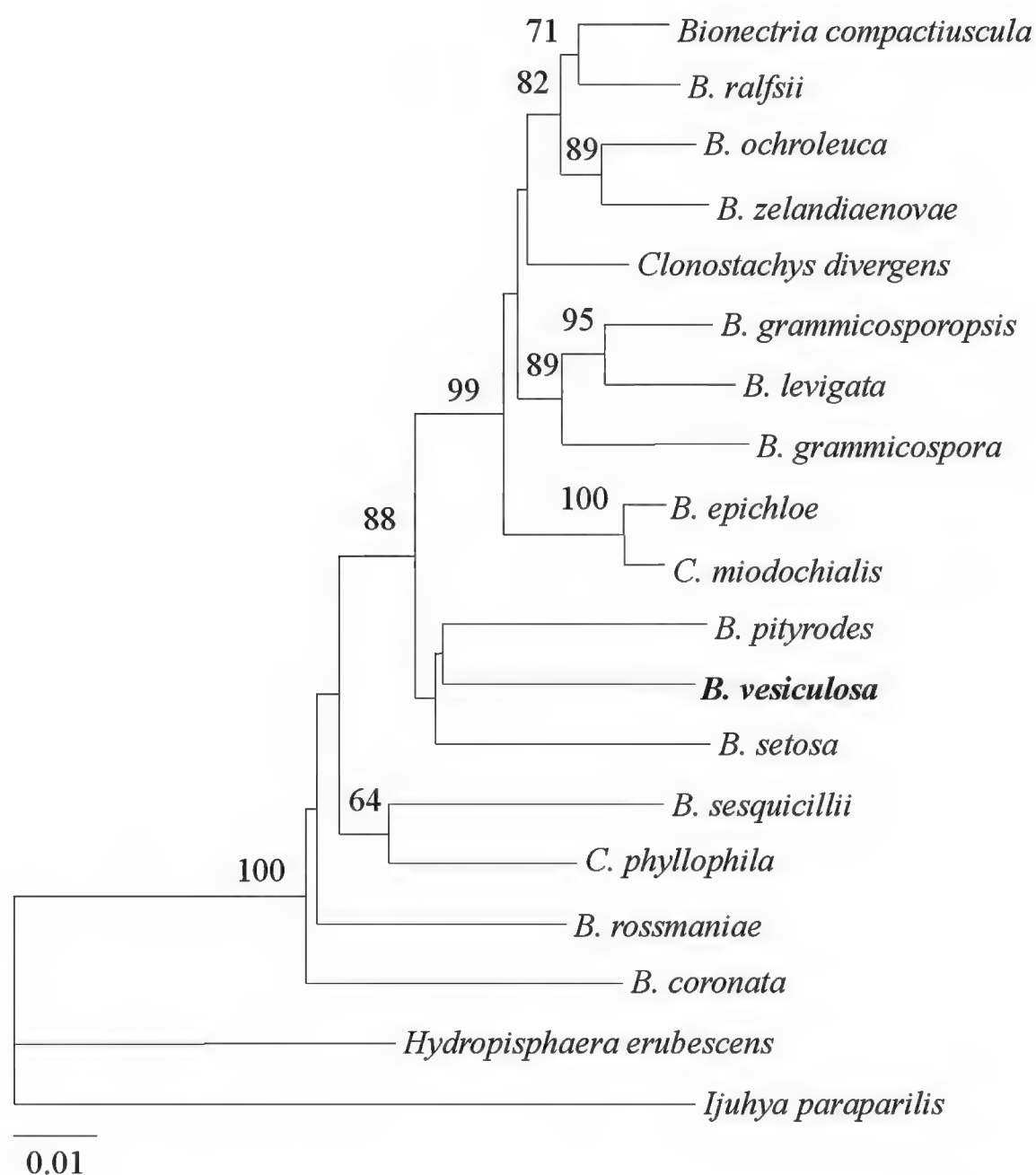


FIG. 3. Neighbour-joining tree based on combined sequences of ITS and 28S nrDNA, showing the relationships among some *Bionectria*/*Clonostachys* species. Bootstrap values  $\geq 50\%$  are noted above internodes.

In conclusion, both morphology and DNA sequence analysis support the recognition of *Bionectria vesiculosa* as a new species.

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## MYCOTAXON

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**Checklist of the arbuscular mycorrhizal fungi (*Glomeromycota*)  
in the Brazilian semiarid**

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**Abstract** — Seventy-nine species of arbuscular mycorrhizal fungi (AMF) are reported for the semiarid Caatinga biome of Northeast Brazil. Data are based primarily on research by L.C. Maia and co-workers during the past 20 years. The full checklist is available at [www.mycotaxon.com/resources/weblists.html](http://www.mycotaxon.com/resources/weblists.html).

**Key words** — *Glomeromycetes*, symbiosis, biodiversity, taxonomy

**Introduction**

Arbuscular mycorrhizal fungi (AMF) form symbiotic association with roots of plants, a mutual connection that may have contributed to the evolution and survival of land-plants and fungi for over 400 million years (Smith & Read 1997).

Thaxter (1922) felt that AMF belonged to the *Endogonaceae*. Based on the symbiotic habit, Morton & Benny (1990) placed all AMF into the new order *Glomales* as a monophyletic group. The AMF are now classified in the phylum *Glomeromycota* (Schüßler et al. 2001) with approximately 220 described species.

Fitter (1990) noted that the fundamental ecological importance of AMF fungi requires research of their diversity in various ecosystems, and in discussing the place of AMF community in a given ecosystem, Sanders et al. (1996) questioned whether there is a relationship between which plants are colonized and what effect AMF have upon both plants and the ecosystem. These questions indicate the need for intensive studies and justify a survey of AMF in different ecosystems.

Most investigations of AMF in Brazil pertain to plant crops and not to natural ecosystems (Maia et al. 2006). The review paper by Trufem (1996) on AMF research within the Amazon, Atlantic Rain Forest, and Cerrado, cited the need for studies in the Caatinga and Pampas, two less studied Brazilian biomes. The Caatinga, which covers more than 800,000 km<sup>2</sup>, representing 70% of the Northeast region and ~11% of Brazil (Drummond et al. 2000), is characterized by a hot dry semiarid climate and vegetation with trees and shrubs (many spiny, some xerophytic) in the *Apocynaceae*, *Bromeliaceae*, *Cactaceae*, *Euphorbiaceae*, and *Leguminosae* (Leal et al. 2003). One recent study (Stürmer & Siqueira 2008) lists only 30 AMF species from the Caatinga biome.

The new records contribute additional data about AMF diversity and a more complete list of AMF species from the Brazilian semiarid Caatinga biome.

### Material and methods

Data cited originated from the authors as well as from the Web of Science; student theses and scientific proceedings have also been considered. References consulted include Albuquerque 2008; Freitas 2006; Lemos 2008; Gattai 2006; Goto et al. 2009, 2010; Lima et al. 2007; Maia et al. 2006; Mergulhão 2007; Mergulhão et al. 2007; Moraes 2007; Pagano et al. 2007; Silva et al. 2007, 2008; and Souza et al. 2007. Gigasporioid-producing AMF species are classified according to Oehl et al. (2008); earlier synonyms are also listed.

### Results and discussion

Seventy-nine species were found in the Caatinga, of which seven are new records for Brazil (*Dentiscutata colliculosa*, *Diversispora spurca*, *Glomus arboreense*, *G. pallidum*, *Racocetra intraornata*, *Scutellospora dipurpureus*, *S. pernambucana*) and three (*D. colliculosa*, *R. intraornata*, *S. pernambucana*) have been recently described. This brings the number of known Brazilian AMF to 106 species, including the 99 taxa cited by Stürmer & Siqueira (2008).

Compared with this last review (Stürmer & Siqueira 2008), the data presented here increase the number of species known in Brazil, which now represents at least 48.2% of the valid species worldwide. Most families of *Glomeromycota* (except *Geosiphonaceae* and *Pacisporaceae*) are represented in the Caatinga, with the number of species representing 74.5% of those recorded from Brazil and 35.9% of those known worldwide.

The majority of AMF studies in the Caatinga have so far focused on agrosystems (Stürmer & Siqueira 2008). However, despite the low number of inventories in the Caatinga, 57 species were listed from vegetation preserved in the biome, almost equaling the number of species (60) recorded from agrosystems throughout Brazil. This preliminary estimate of the AMF diversity

in the Caatinga suggests that a high diversity will probably be found in the biome in the future, particularly considering the high number of plants and animals also present (Leal et al. 2003).

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## MYCOTAXON

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**A new species of *Spadicoides* from Yunnan, China**

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**Abstract** — *Spadicoides yunnanensis* sp. nov., collected from tropical forests in Yunnan province of China, is described and illustrated from a specimen occurring on dead branches of *Camellia japonica*.

**Key words** — hyphomycetes, taxonomy

**Introduction**

The genus *Spadicoides* was established by Hughes (1958) with *S. bina* (Corda) S. Hughes as the type species. Sinclair et al. (1985) amended the generic description to include species with solitary conidia on branched or unbranched conidiophores and suggested that the production of conidia in chains is the sole diagnostic character separating *Diplococcium* Grove from *Spadicoides*. Goh & Hyde (1996) reviewed the genus *Spadicoides* and recognized 21 species in this genus. The genus is characterized by differentiated, single, unbranched or branched conidiophores with polytretic, terminal and intercalary conidiogenous cells producing solitary, terminal and lateral, euseptate, obovoid to ellipsoid conidia (Ellis 1971; Kuthubutheen & Nawawi 1991). Thus far, 30 species have been accepted in *Spadicoides*, of which four are described from China (Zhou et al. 1999; Ho et al. 2002; Wong et al. 2002; Cai et al. 2004).

Most species of *Spadicoides* are saprobes on rotten leaves or dead branches. A continuing survey of saprobic fungi on dead wood from tropical forest in Yunnan province of China revealed a previously undescribed species, *Spadicoides yunnanensis*. The type specimen is deposited in HSAUP (Herbarium of the Department of Plant Pathology, Shandong Agricultural University) with isotypes in HMAS (Mycological Herbarium, Institute of Microbiology, Chinese Academy of Sciences).

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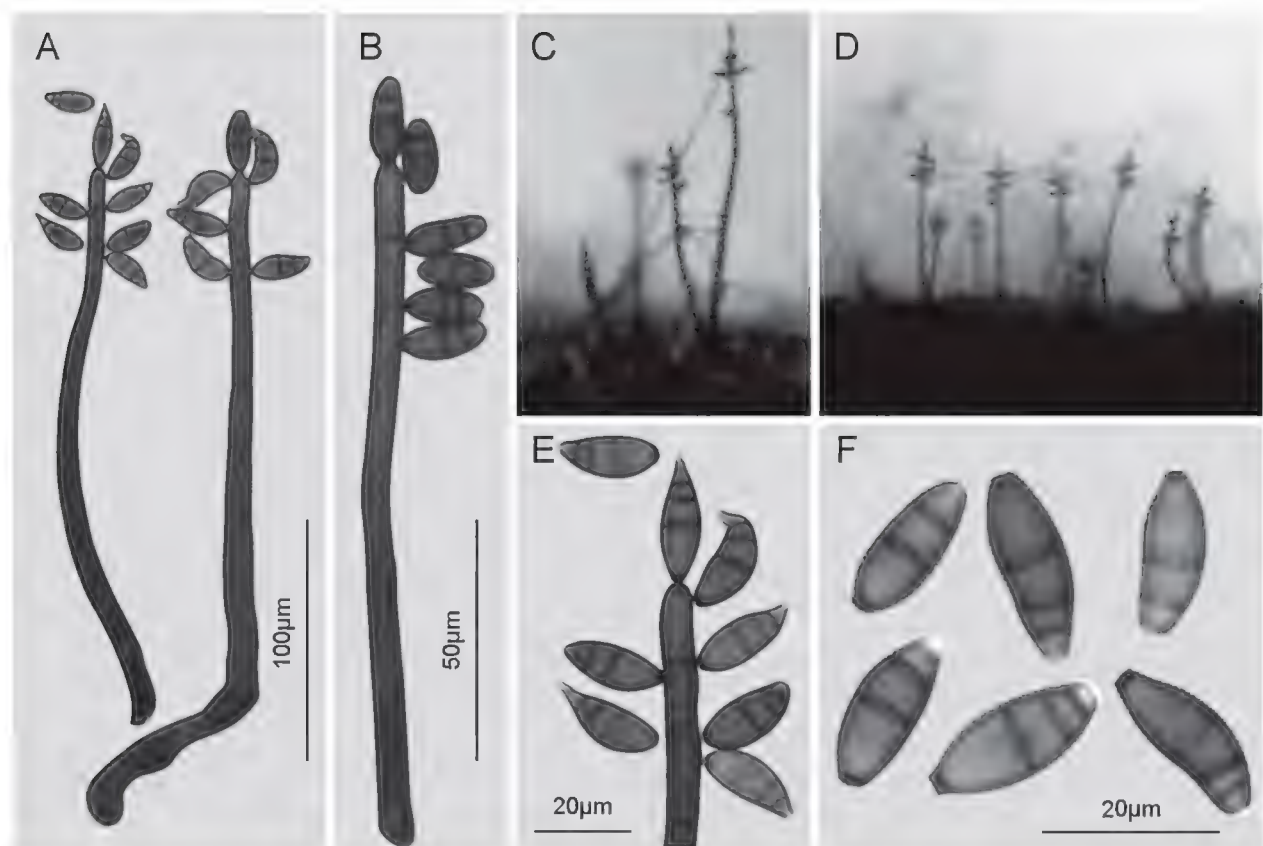


FIG. 1. *Spadicoides yunnanensis*. A–B. Conidiophores with terminal and lateral conidia. C–D. Conidiophores arising from wood. E. Conidiogenous cells showing conidiogenous pores. F. Conidia with 2–3 eusepta.

Taxonomic description

*Spadicoides yunnanensis* L.G. Ma & X.G. Zhang, sp. nov.

FIGURE 1

MYCOBANK MB 518363

*Coloniae effusae in substrato naturali, atro-brunneae. Mycelium partim superficiale, partim immersum, ex hyphis ramosis, septatis, pallide brunneis, laevibus, 1.5–2.0 µm crassis compositum. Conidiophora macronematosa, mononematosa, singula, simplicia, non ramosa, erecta, cylindrica, recta vel flexuosa, laevia, atro-brunnea, 7–12-septata, 140–290 µm longa, ad basim 10.0–14.5 µm crassa, ad apicem 6.5–8.0 µm crassa. Cellulae conidiogenae polytreticae, in conidiophoris incorporatae, terminales et intercalares, brunnea. Conidia solitaria, acropleurogena, simplicia, obpyriformia vel ovoidea, ad apicem rotundata vel acuta, ad basim truncata, brunnea, interdum cellulae apicalis subhyalina, laevia, crassitunicata, 2–3-euseptata, 18.5–28.0 µm longa, 6.5–10.0 µm crassa. Basi truncata 2.0–3.5 µm lata.*

HOLOTYPE: on dead branches of *Camellia japonica* L. (*Theaceae*), the Forbidden Forest of Banna, Yunnan Province, China. Oct. 16. 2008, L.G. Ma, HSAUP H0041 (Isotype HMAS 196882).

ETYMOLOGY: in reference to the province where the type was found

Colonies effuse on natural substratum, dark brown. Mycelium partly superficial, partly immersed, composed of branched, septate, pale brown, smooth-walled hyphae, 1.5–2.0 µm thick. Conidiophores macronematous, mononematous,

single, simple, unbranched, erect, cylindrical, straight or flexuous, smooth, dark brown, 7–12-septate, 140–290 µm long, 10.0–14.5 µm wide at the base, 6.5–8.0 µm wide at the apex. Conidiogenous cells polytretic, integrated, terminal and intercalary, brown. Conidia solitary, acropleurogenous, simple, obpyriform to ovoid, apex rounded or acute, base truncate, brown, occasionally apical cell subhyaline, smooth-walled, thick-walled, 2–3-euseptate, 18.5–28.0 µm long, 6.5–10.0 µm wide in the broadest part, 2.0–3.5 µm wide at the truncate base.

The conidia of *S. yunnanensis* resemble those of *S. curvularioides* (Sutton 1978) and *S. xylogena* (Hughes 1958) in having a similar conidial size and number of septa. However, the conidia of *S. curvularioides* are verrucose, pale brown, and cymbiform compared to the smooth, brown, obpyriform to ovoid conidia of *S. yunnanensis*, and the apices of the conidiophores in *S. curvularioides* are geniculate as opposed to those of *S. yunnanensis*, which are not. In addition, *S. yunnanensis* can be separated from *S. xylogena* by its obpyriform to ovoid conidia without banded septa.

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## MYCOTAXON

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**New species of *Monodictys* and *Veronaea* from soil  
in the Yellow River source area, China**HAO-QIN PAN<sup>1, 2</sup> & TIAN-YU ZHANG<sup>1\*</sup>

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**Abstract**—Two new species, *Monodictys macrospora* and *Veronaea latispora* found from wetland soil and Gobi soil in Yellow River source area of China are described, illustrated and compared with similar taxa. The type specimens (dried cultures) and living cultures are deposited in the Herbarium of Shandong Agricultural University Plant Pathology (HSAUP). Isotypes are kept in the Herbarium of Institute of Microbiology, Academia Sinica (HMAS).

**Key words**—dematiaceous hyphomycetes, taxonomy, soil fungi

**Introduction**

During an investigation of soil dematiaceous hyphomycetes in Yellow River source area, China, two species in the genera *Monodictys* S. Hughes and *Veronaea* Cif. & Montemart. were discovered. Based on their distinctive morphological characteristics they could not be assigned to any of the described species. They are treated as new species and are compared to the most closely related species, *Monodictys chlamydosporoidea* (Liu & Zhang 2007), *M. arxanensis* (Wu & Zhang 2008), *Veronaea parvispora* (Ellis 1976), and *V. musae* (Ellis 1976).

**Taxonomy*****Monodictys macrospora* H.Q. Pan & T.Y. Zhang, sp. nov.**

FIG. 1

MYCOBANK MB 512616

*Coloniae in PCA effusae, atrobrunneae vel atrae. Mycelium maximam parten superficiale et aliquot immersum, ex hyphis modice vel atrobrunneis, levibus, septatis, 5–6.5 µm crassis compositum. Conidiophora micronematica, recta vel curvata, pallide brunnea,*

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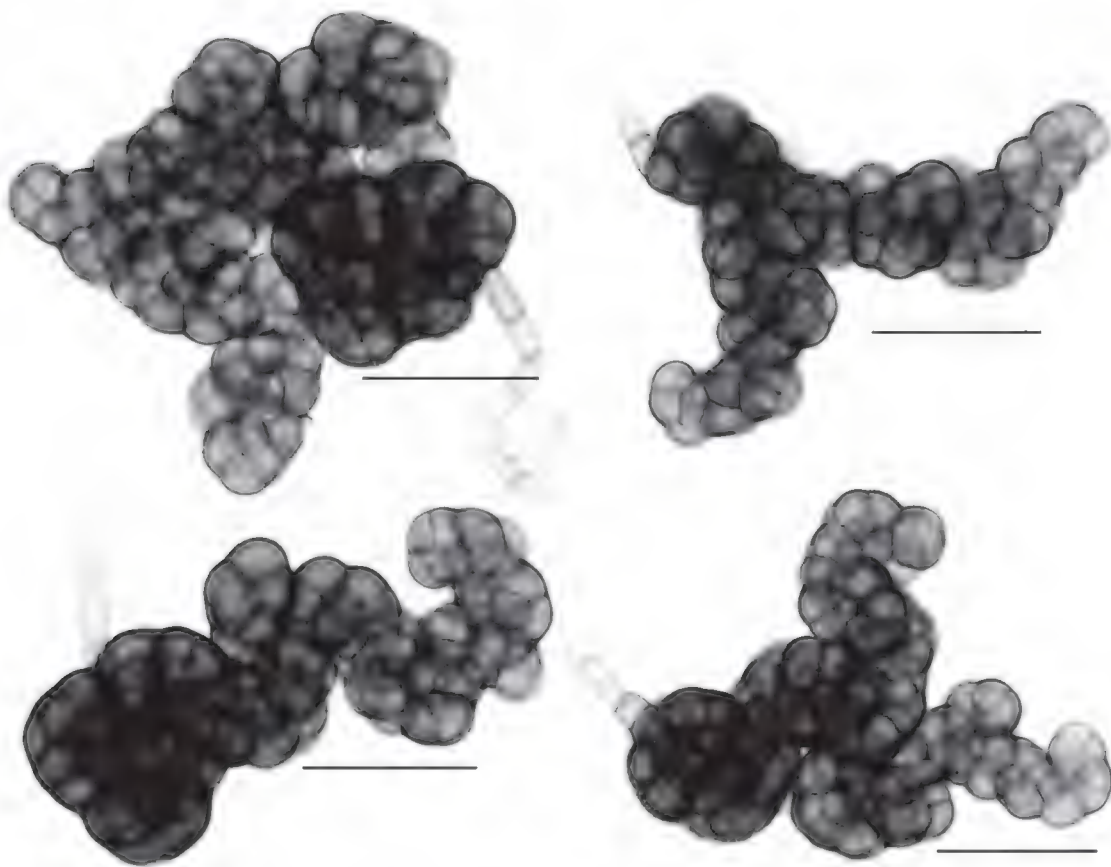


FIG. 1 Conidia and conidiophores of *Monodictys macrospora*  
(ex holotype; bars = 100  $\mu\text{m}$ )

*laevia*. Cellulae conidiogenae pallide brunneae, monoblasticae, leaves, determinatae, aliquando inflatae, subglobosae vel cylindricae, 10.5–28.5  $\mu\text{m}$  longae et 6.5–13  $\mu\text{m}$  crassae. Conidia solitaria, acropleurogena, irregularis, ex cellulis globosis numerosis crasse inflatis composite, brunnea vel atrobrunnea, 43–125  $\times$  21–38  $\mu\text{m}$ .

HOLOTYPE: isolated from wetland soil, Madoi County, Qinghai Province, China, 24 Jul. 2007, H.Q. Pan, HSAUP II<sub>07</sub>4068, holotype, HMAS196216, isotype.

ETYMOLOGY: in reference to the large conidia.

Colonies in potato carrot agar medium effuse, dark brown to black. Mycelium mostly superficial. Hyphae moderate brown to dark brown, smooth, septate, branched, 5–6.5  $\mu\text{m}$  wide. Conidiophores micronematous, straight or flexuous, pale brown, smooth. Conidiogenous cells pale brown, monoblastic, smooth, determinate, sometimes inflated, subglobose or cylindrical, 10.5–28.5  $\mu\text{m}$  long, 6.5–13  $\mu\text{m}$  wide. Conidia solitary, acropleurogenous, irregular, comprising numerous globose inflated cells, brown to dark brown, 43–125  $\times$  21–38  $\mu\text{m}$ .

This species is similar to *Monodictys chlamydosporoidea* H.M. Liu & T.Y. Zhang and *Monodictys arxanensis* Y.M. Wu & T.Y. Zhang in conidial morphology. Both *M. chlamydosporoidea* and *M. arxanensis* have smaller conidia; those of *M. chlamydosporoidea* are 23–44  $\times$  17–30  $\mu\text{m}$  and those of *M. arxanensis* are 25–60  $\times$  2–25  $\mu\text{m}$ . These two species also have relatively simple conidia, pale brown colonies, and hyaline conidiophores.

***Veronaea latispora*** H.Q. Pan & T.Y. Zhang, sp. nov.

FIG. 2

MYCOBANK MB 512622

*Coloniae effusae, olivaceae. Conidiophora recta vel leviter curvata, septata, brunnea, laevia, usque ad 90 µm longa, 1.5–2.5 µm crassa, apicem versus cicatricibus conidialibus minutis numerosis praedita. Conidia late obovoidea, subhyalina vel pallide brunnea, laevia, 7.5–9.5 × 3–5 µm.*

HOLOTYPE: isolated from Gobi soil, Xunhua County, Qinghai Province, China. 24 Aug 2006, H.Q. Pan, HSAUP II<sub>06</sub>3223, holotype; HMAS196217, isotype.

ETYMOLOGY: in reference to the relatively broad conidia of this species.

Colonies effuse, olivaceous brown. Conidiophores straight or slightly curved, septate, brown, smooth, up to 90 µm long, 1.5–2.5 µm thick, with numerous minute scars at the upper parts. Conidia broadly obovoid, subhyaline to pale brown, smooth, 7.5–9.5 × 3–5 µm.

The most closely related species in conidial morphology to this new taxon are *Veronaea parvispora* M.B. Ellis and *V. musae* M.B. Ellis (Ellis 1976). However, the conidia of *V. parvispora* are much smaller (2–3 × 1.5–2 µm). *Veronaea musae* differs from the new taxon in having narrower conidia (5–10 × 2–3 µm), usually with a minutely papillate base.

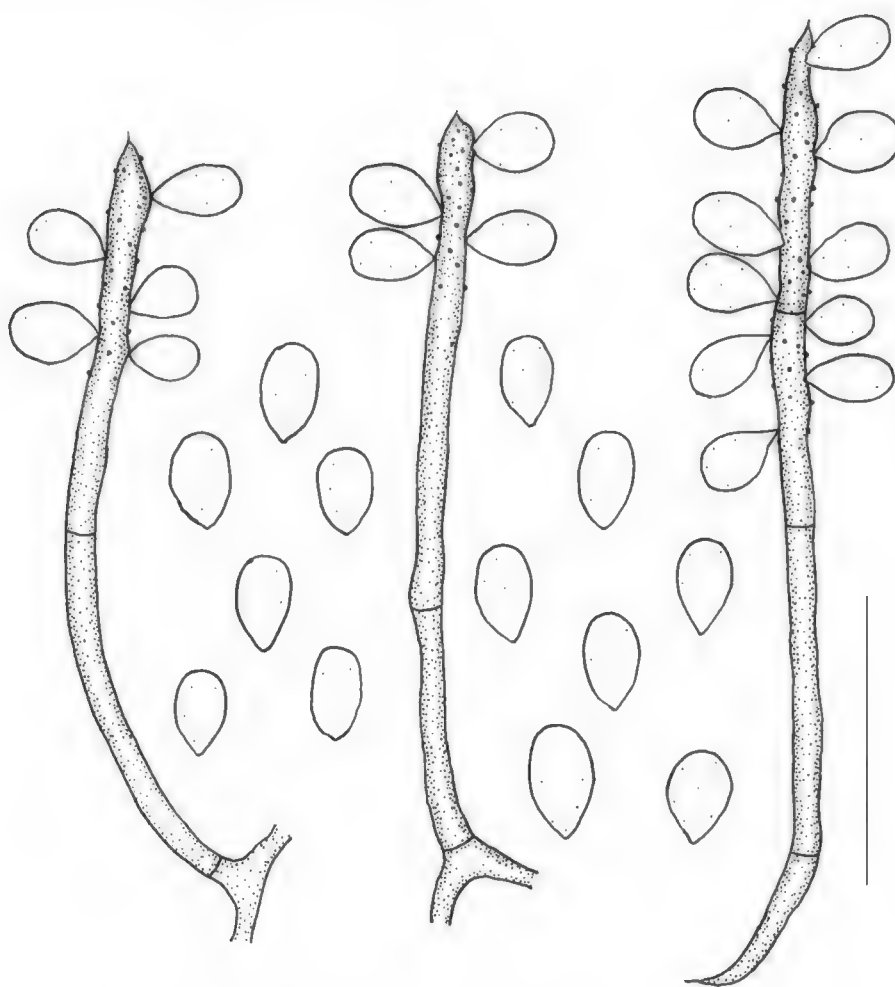


FIG. 2 Conidia and conidiophores of *Veronaea latispora* (ex holotype; bars = 25 µm)

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## A new *Trametes* species from Southwest China

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**Abstract** — A new polypore, *Trametes cystidiolophora* sp. nov., found in Yunnan Province, southwest China, is described and illustrated. The new species is characterized by its pale grayish brown to pale cinnamon-buff pileus with distinctly concentric zones and radial veins, uneven pore surface, cylindrical to more or less allantoid basidiospores ( $6.6\text{--}9.2 \times 2.4\text{--}3 \mu\text{m}$ ), abundant cystidioles present in the hymenium, and skeletal-binding hyphae that become swollen in KOH.

**Key words** — *Polyporaceae*, taxonomy, wood-rotting fungi

### Introduction

The genus *Trametes* Fr. is characterized by having pileate basidiocarps, a trimitic hyphal system with clamp connections on generative hyphae, hyaline and thin-walled basidiospores that are negative in Melzer's reagent, and causing white rot (Gilbertson & Ryvarden 1986, Ryvarden & Gilbertson 1994, Lindblad & Ryvarden 1999, Núñez & Ryvarden 2001). About 50 species in the genus have been reported in the world (Kirk et al. 2008), including 23 species previously recorded from China (Zhao & Zhang 1991, Teng 1996, Zhao 1998, Dai et al. 2007, Dai 2009, Dai & Yuan 2010).

During the study on wood-decaying fungi from Gaoligongshan Nature Reserve, Yunnan Province, southwest China, a species of *Trametes* was found that could not be identified to any known species. It is described in the present paper as *Trametes cystidiolophora*.

### Materials and methods

The studied specimens are deposited in herbaria as cited below. The microscopic procedure follows Dai & Penttilä (2006). Sections were studied at

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magnification up to  $\times 1000$  using a Nikon Eclipse E 80i microscope and phase contrast illumination. Drawings were made with the aid of a drawing tube. To present spore size variation, the 5% of measurements excluded from each end of the range are given in parentheses. Abbreviations include IKI = Melzer's reagent, IKI- = negative in Melzer's reagent, KOH = 5% potassium hydroxide, CB = Cotton Blue, CB- = acyanophilous, L = mean spore length (arithmetic average of all spores), W = mean spore width (arithmetic average of all spores), Q = variation in the L/W ratios between the specimens studied, and n = number of spores measured from given number of specimens. Special colour terms follow Anonymous (1969) and Petersen (1996).

### Taxonomy

*Trametes cystidiolophora* B.K. Cui & H.J. Li, sp. nov.

FIG. 1

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*Carpophorum annuum*, *pileatum*, *imbricatum*; *facies pororum bubalina vel reseobubalina*, *pori rotundi vel angulati*, 2–3 per mm. *Systema hypharum trimiticum*, *hyphae generatoriae hyalinae, fibulatae, hyphae skeletales contexti* 2.8–6.2  $\mu\text{m}$ ; *spores hyalinae, cylindricae, IKI-, CB-,* 6.6–9.2  $\times$  2.4–3  $\mu\text{m}$ .

TYPE — China. Yunnan Province, Baoshan, Gaoligongshan Nature Reserve, on dead angiosperm tree, 25.X.2009 Cui 8084 (holotype in BJFC); Cui 8087 (isotype in BJFC).

ETYMOLOGY — *cystidiolophora* (Greek): = “cystidiole-bearing”, referring to the abundant cystidioles in the hymenium.

FRUITBODY — Basidiocarps annual, pileate, usually imbricate, without odor or taste when fresh, corky and light in weight when dry. Pileus dimidiate to semicircular, projecting up to 4.2 cm, 7.3 cm wide, 7 mm thick at the base; pileal surface pale grayish brown to pale cinnamon-buff when dry, glabrous, distinctly concentrically zoned and radially veined; margin sharp, wavy or incised in rounded lobes, deflexed with age. Pore surface cream-buff to pinkish buff when dry, slightly shiny; sterile margin white to cream, up to 2.5 mm wide; pores round to angular, 2–3 per mm, dissepiments thin, entire at margin and dentate to hydroid with age. Context cream, corky, up to 3 mm thick. Tubes cream to cream-buff, corky, up to 4 mm long.

HYPHAL STRUCTURE — Hyphal system trimitic; generative hyphae with clamp connections; skeleto-binding hyphae dominant, thick-walled to subsolid, IKI-, CB-, become swollen in KOH.

CONTEXT — Generative hyphae infrequent, hyaline, thin-walled, moderately branched, 2–3.7  $\mu\text{m}$  in diam; skeletal hyphae dominant, hyaline, slightly thick-walled to subsolid, frequently branched, and the slightly thick-walled skeletal hyphae often collapsed, interwoven, 2.8–6.2  $\mu\text{m}$  in diam; binding hyphae hyaline, thick-walled to almost solid, frequently branched, interwoven, 1.7–3  $\mu\text{m}$ .

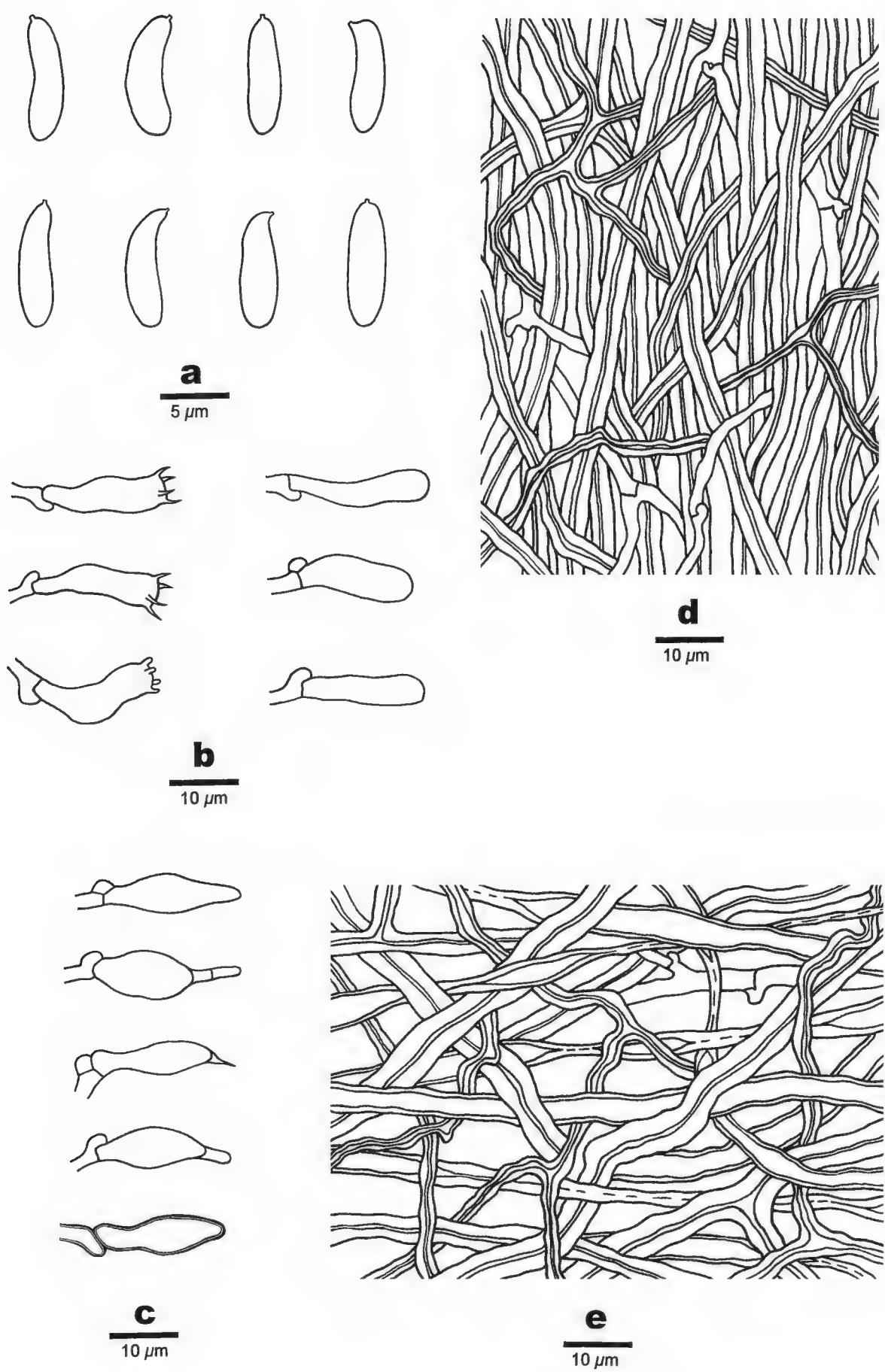


FIG. 1. Microscopic structures of *Trametes cystidiolophora* (drawn from the holotype).  
a: Basidiospores. b: Basidia and basidioles. c: Cystidioles.  
d: Hyphae from tube trama. e: Hyphae from context.

**TUBES** — Generative hyphae infrequent, hyaline, thin-walled, frequently branched, 1.7–3  $\mu\text{m}$  in diam; skeletal hyphae dominant, hyaline, thick-walled to subsolid, occasionally branched, interwoven, 2.3–5  $\mu\text{m}$ ; binding hyphae hyaline, flexuous, thick-walled to almost solid, frequently branched, interwoven, 1.6–3.1  $\mu\text{m}$ . Cystidia absent, cystidioles abundant in the hymenium, fusoid, hyaline, mostly thin-walled, occasionally slightly thick-walled, some with one or two septa, 16–24  $\times$  4–6  $\mu\text{m}$ ; basidia clavate, with four sterigmata and a basal clamp connection, 16–18.2  $\times$  5–7.8  $\mu\text{m}$ ; basidioles in shape similar to basidia, but slightly smaller.

**SPORES** — Basidiospores cylindrical, occasionally slightly curved to more or less allantoid hyaline, thin-walled, smooth, IKI–, CB–, (6–)6.6–9.2(–10)  $\times$  (2.2–)2.4–3(–3.3)  $\mu\text{m}$ , L = 8.1  $\mu\text{m}$ , W = 2.79  $\mu\text{m}$ , Q = 2.78–3.04 (n=60/2).

**TYPE OF ROT** — White rot.

**REMARKS** — *Trametes cystidiolophora* is characterized by its pale grayish brown to pale cinnamon-buff pileal surface with distinctly concentric zones and radial veins, uneven pore surface, cylindrical to more or less allantoid basidiospores (6.6–9.2  $\times$  2.4–3  $\mu\text{m}$ ), and abundant cystidioles present in the hymenium. Moreover, its skeleto-binding hyphae become swollen in KOH.

*Trametes cystidiolophora* may be confused with *Fomitopsis palustris* (Berk. & M.A. Curtis) Gilb. & Ryvarden, which produces similar basidiospores, but the two species can be separated by the rot type. *Trametes cystidiolophora* causes a white rot, while *Fomitopsis palustris* causes a brown rot.

*Trametes maxima* (Mont.) A. David & Rajchenb. is similar to *T. cystidiolophora* by sharing similar uneven pore surface and pore size (2–3 per mm), but *T. maxima* differs in its tomentose to hirsute pileal surface and smaller basidiospores (4.5–5.5  $\times$  2–2.5  $\mu\text{m}$ , Gilbertson & Ryvarden 1986).

*Trametes cotonea* (Pat. & Har.) Ryvarden, which has similar basidiospores (7–10  $\times$  2.5–3.5  $\mu\text{m}$ ) as *T. cystidiolophora*, is usually effused-reflexed, paper thin, and flexible and its pores are smaller (3–4 per mm, Ryvarden & Johansen 1980).

*Trametes glabrata* (Lloyd) Ryvarden, which shares with *T. cystidiolophora* a glabrous pileus, uneven pore surface, and similar pore size. However, *T. glabrata* has distinctly smaller basidiospores (4–5  $\times$  1–1.5  $\mu\text{m}$ , Ryvarden 1992).

Cystidioles are also present in several other reported species in *Trametes*, such as *T. gibbosa*, *T. hirsuta*, *T. ljubarskyi*, and *T. pubescens* (Gilbertson & Ryvarden 1986, Núñez & Ryvarden 2001), but the cystidioles in all these species are infrequent and without septa. The fact that its cystidioles are abundant in the hymenium and that some are septate make *T. cystidiolophora* unique in *Trametes*.



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## MYCOTAXON

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**First description of *Oidium neolycopersici* (Erysiphaceae) in France, on a new host plant extinct in the wild**DAVID DELMAIL<sup>1,2\*</sup> & JEAN-LUC AUTRET<sup>2</sup>

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**Abstract** — The first description of *Oidium neolycopersici* (Erysiphaceae) discovered on a Madeiran plant now extinct in the wild, *Normania triphylla* (Solanaceae), is provided. The pathogen was collected from the National Botanical Conservatory of Brest in western France.

**Key words** — Erysiphales, mildew, Solanales, Madeira

*Normania triphylla* Lowe plants (Solanaceae) cultivated in the greenhouse at the National Botanical Conservatory of Brest (NBCB) showed signs of powdery mildew. This Madeiran plant is extinct in the wild and currently the only ex situ culture exists at the NBCB. Recently several dense and discontinuous white patches were observed on the leaf upper epidermis of 47% of individuals. These characteristics were easily differentiated from symptoms caused by *Leveillula taurica*, which can readily affect Solanaceae, a fungus considered to be the unique agent of powdery mildew on *N. triphylla* in France up to now that causes white powdery masses appearing just under the chlorotic spots that are produced on the adaxial leaf surface.

To determine the morphological characteristics of the pathogen affecting *N. triphylla*, the surface mycelium was removed with adhesive tape and examined under optical microscope. Microscopic observations revealed exclusively solitary ellipsoid conidia ( $29.3 \times 13.5 \mu\text{m}$ ) germinating with one short germ tube terminating in simple apices (FIG. 1A). Conidiophores were straight, with cylindrical foot cells ( $43.0 \times 9.3 \mu\text{m}$ ), sometimes followed by a longer cell and one or two shorter cells (FIG. 1B). Fibrosin bodies and chasmothecia were not observed. Based on these characteristics the fungus was

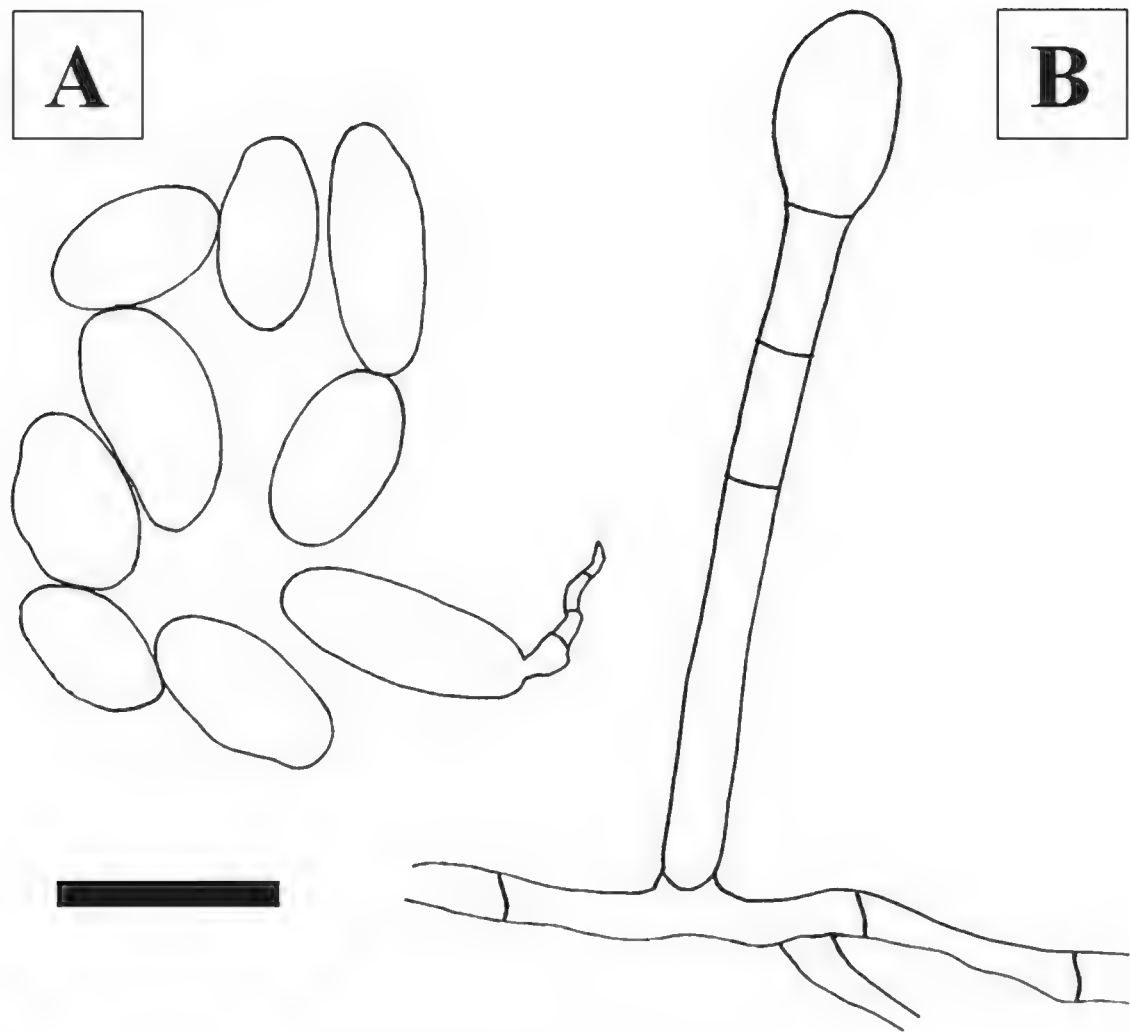


FIG. 1 A–B. Drawings of *Oidium neolycopersici* specimens observed on *Normania triphylla*. A. Free conidia with a germinating conidium. B. Conidium developing singly on conidiophore. Scale bar represents 30  $\mu\text{m}$ .

identified as *Oidium neolycopersici* L. Kiss (Kiss et al. 2001). This species has been reported as occurring on host plants in the *Solanaceae* (*Solanum betaceum* and *S. lycopersicum*) family in Asia (Baiswar et al. 2009; Kiss et al. 2001; Li et al. 2008, Yolageldi et al. 2008), Australia, Tanzania, the French Caribbean (Kiss et al. 2001), North America (Kiss et al. 2001; Kiss et al. 2005), and Europe (Ivic et al. 2009; Kiss et al. 2001). Specimens were identified on tomato in West Europe by sequencing by Kiss et al. (2001) in France (specimen examined: BPI 747013; database accession number: AF229019) and Netherlands (specimen examined: VPRI 20724; database accession number: AF229015).

To confirm the pathogenicity, 15 healthy *N. triphylla* plants were inoculated with conidia from infected plants and then kept in a polypropylene (PP) chamber placed in a greenhouse cabinet at  $25 \pm 1^\circ\text{C}$  and a 15-h photoperiod for 7 days. The PP chamber was then removed and plants grown in the greenhouse. After 9 days, powdery mildew symptoms appeared on the inoculated leaves of the plants and the morphological characteristics of the reisolated pathogen were the same as those observed on the naturally infected plants.



This is the first report of powdery mildew caused by *O. neolycopersici* on *N. triphylla* in France. This disease has the potential to be extremely virulent (Jones et al. 2001) and may become a problem in ex situ cultures of *N. triphylla*. These cultures are essential because reintroduction attempts, which have failed until now, will help researchers learn how *N. triphylla* might again grow wild in the laurel forest of Madeira.

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## MYCOTAXON

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**Some new pyrenomycetous and loculoascomycetous fungi on the endemic Hawaiian plant *Hibiscadelphus giffardianus***LARISSA N. VASILYEVA<sup>1</sup> & JACK D. ROGERS<sup>2</sup>*vasilyeva@biosoil.ru & rogers@wsu.edu*<sup>1</sup>*Institute of Biology & Soil Science, Far East Branch of the Russian Academy of Sciences, Vladivostok 690022, Russia*<sup>2</sup>*Department of Plant Pathology, Washington State University Pullman, WA 99164-6430, USA*

**Abstract** — The fungal associates of the rare tree *Hibiscadelphus giffardianus* were studied. Three of these (*Eutypella giffardiani*, *Thyridaria hawaiiensis*, and *Valsonectria macrospora*) are described and illustrated as new to science.

**Key words** — *Ascomycota*, Hawai'i, taxonomy

**Introduction**

Several unknown pyrenomycetous and loculomycetous fungi inhabiting dead branches of *Hibiscadelphus giffardianus* Rock (a Hawaiian endemic in the *Malvaceae*) were collected by J.D. Rogers in 2005; three of these species are described below. The Hawaiian Islands have the very high (90%) degree of endemism associated with an exceptionally diverse flora (Kim et al. 1998). For the mushrooms, 46 of 310 species are endemic Hawaiian taxa (Hemmes & Desjardin 2002). Other fungal groups (*Capnodiales*, *Chaetothyriales*, *Coronophorales*, *Corticiales*, *Diaporthales*, *Diversisporales*, *Dothideales*, *Erysiphales*, *Helotiales*, *Hymenochaetales*, *Meliolales*, *Microthyriales*, *Myriangiales*, *Phyllachorales*, *Pleosporales*, *Uredinales*, *Xylariales*) also have endemic Hawaiian representatives (Stevens 1925; Petrak 1952, 1953; Goos 1970; Sutton & Hodges 1983; Hodges & Gardner 1984; Hodges 1985; Barr & Hodges 1987; Gardner 1988, 1990, 1996; Goos & Uecker 1992; Koske & Gemma 1995; Gilbertson & Nakasone 2003; Rogers et al. 2003, 2006, 2007, 2008; Scholler & Aime 2006). We are not aware of any other reports of fungi on *Hibiscadelphus*. This paper contributes to a better understanding of the uniqueness of Hawaiian mycobiota.

## Materials and methods

Microscopic analyses were carried out using standard techniques. Observations, measurements, and photographs of asci and ascospores were made using Zeiss Primo Star and Leica MZ75 microscopes, G10 and Canon Power Shot S40 digital cameras, as well as AxioVision software. Photographs of stromata were taken using a Nikon D40x digital camera. Measurements of asci and ascospores were made in water mounts. Colors follow those of Rayner (1970).

## Taxonomic descriptions

*Eutypella giffardiani* Lar.N. Vassiljeva & J.D. Rogers, sp. nov.

FIGS. 1A–B

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*Stromata cortice erumpentia, valsoidea, gregaria, parte immersa nigro limitata, circular vel ellipsoidea, disco ostiolato nigro, 1–2 mm diam., praedita. Perithecia profunde immersa, monosticha, globosa, 300–350 µm diam.; collis leniter elongatis, sulcatis, 230–250 µm diam. Asci fasciculati, paraphysati, unitunicati, octospori, clavati vel fusoides, membrana apicem versus incrassata, annulo apicali in liquore iodato Melzeri cyanescente, partibus sporiferis 25–35 × 4.5–5 µm, stipitibus 15–25 µm longitudine. Ascosporae unicellulares, allantoidae, irregulariter biseriatae vel conglobatae, subolivaceae, 6–8 × 1.7–2 µm.*

**HOLOTYPE:** Hawai'i, Island of Hawaii, Hawaii Volcanoes National Park, Kipuka Puauulu (Bird Park), dead branches of *Hibiscadelphus giffardianus*, 3 November 2005, Jack D. Rogers (BISH). **Isotype:** VLA.

**STROMATA** erumpent through the bark, valloid, aggregated, bounded internally by a black zone line, circular to elliptical, with ostiolar disc 1–2 mm diam., surface black. **PERITHECIA** deeply embedded, globose, 300–350 µm diam.; perithecial necks somewhat elongate, sulcate, 230–250 µm diam. **ASCI** in paraphysate fascicles, unitunicate, eight-spored, clavate or spindle-shaped, with tiny amyloid apical ring, p. sp. 25–35 × 4.5–5 µm, with stipes 15–25 µm long. **ASCOSPORES** one-celled, overlapping and irregularly biseriate or crowded, allantoid, subolivaceous, 6–8 × 1.7–2 µm.

**COMMENTS**—Numerous species of *Eutypella* have been described. They are difficult to differentiate, and the identification key by Rappaz (1987) offers the most useful information. Two ranges of ascospore average length (5–8 µm and 7–11 µm), which are repeated many times in the key, characterize large groups in the genus. The specimen from Hawaii falls into the group with smaller size range. The ascospore width correlates well with length, and almost all species with ascospores 5–8 µm long are narrower than 2 µm, whereas species with an ascospore length of 7–11 µm have ascospores wider than 2 µm (most often 2–2.5 µm).

As there are 21 species with an ascospore length of 5–8 µm (Rappaz 1987), it is important to find other differences to distinguish between them. Two species [*E. comosa* (Speg.) Rappaz, *E. leucaenae* Rehm] are rare exceptions that can



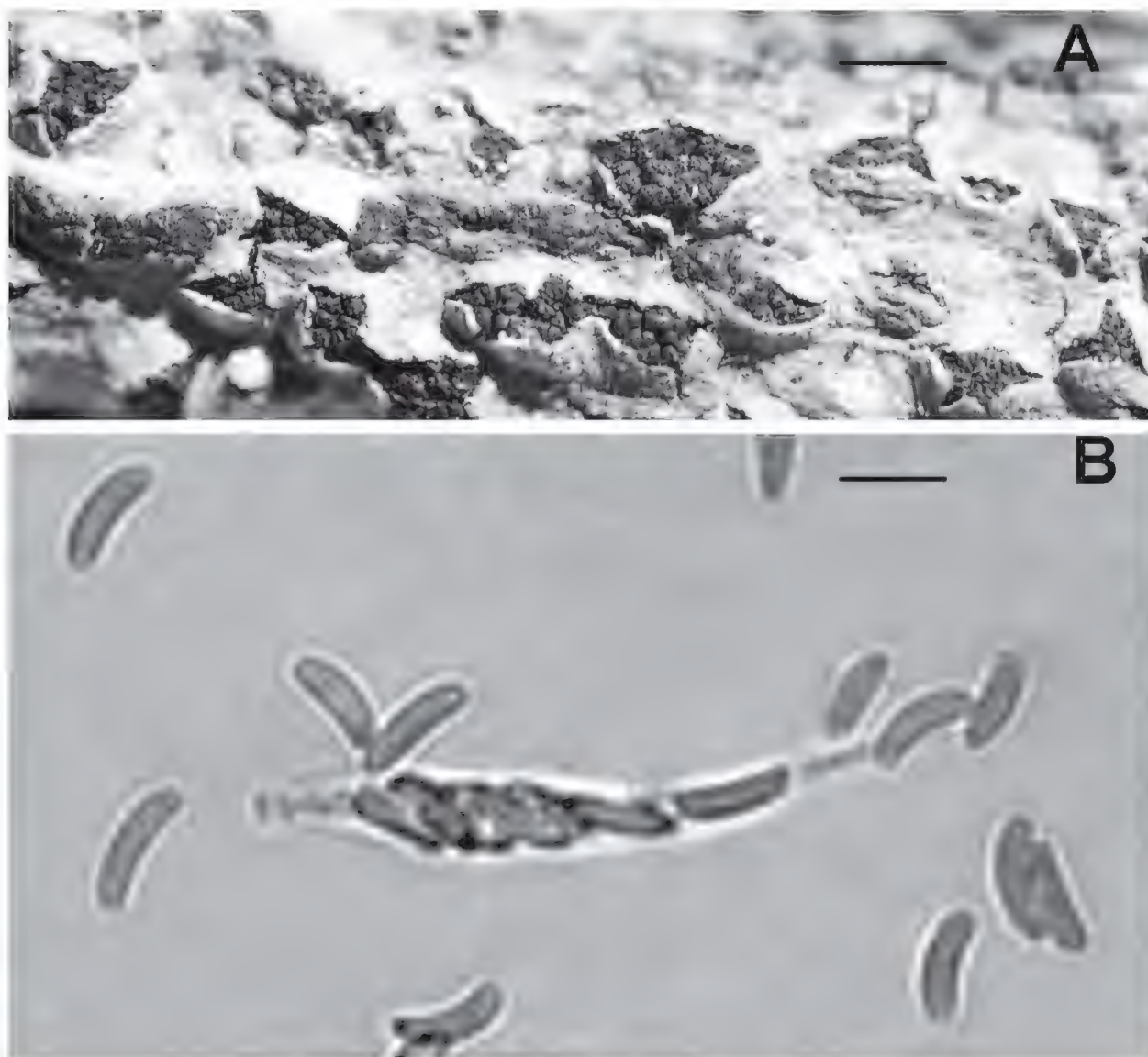


FIG. 1. *Eutypella giffardiani*. A. Stromata. B. Ascus and ascospores.  
Scale bars: A = 1 mm; B = 6  $\mu$ m.

be easily separated by ascospores that are more than 2  $\mu$ m broad, and another species, *E. hunanensis* Rappaz, produces very narrow ascospores less than 1.2  $\mu$ m wide. Six species [*E. arecae* (Syd. & P. Syd.) Rappaz, *E. bonariensis* Speg., *E. gliricidae* Rehm, *E. prunastri* (Pers.) Sacc., *E. sorbi* (Alb. & Schwein.) Sacc., *E. theobromicola* Wakef.] display a J-negative reaction of the ascial apical ring that contrasts with a J-positive one in the Hawaiian specimen.

Another character differentiating species of *Eutypella* is the size of the ostioles. *Eutypella androssowii* Rehm and *E. tetraploa* (Berk. & M.A. Curtis) Sacc. have ostioles with a diam of 100–150  $\mu$ m and 100–180  $\mu$ m, respectively, whereas 200  $\mu$ m diam ostioles occur in *E. andicola* Speg., *E. atropae* (Mont.) Sacc., and *E. capensis* Rappaz; 150–200  $\mu$ m diam ostioles are observed in *E. padina* (Nitschke) Nannf. and *E. sarcobati* Ellis & Everh. Ostioles in *E. extensa* (Fr.) Sacc. are 180–220  $\mu$ m diam, compared to a diameter ~220–250  $\mu$ m in the Hawaiian specimen. As such, they correspond to *E. alsophila* (Durieu & Mont.) Berk. in the group with the range in ascospore length of 5–8  $\mu$ m. However,

*E. alsophila* falls into the group with 400–600 µm diam perithecia, not the group with 200–400 µm diam perithecia.

The Hawaiian specimen belongs to the group with 200–400 µm diam and the combination of the other diagnostic features — ascospore, ostiole, and perithecial sizes and the J-positive apical ring — corresponds to the sole remaining species, *E. kochiana* Rehm. There is no information about ostiolar size in *E. kochiana*, but it has smaller ascospores than the Hawaiian specimen. Although Rappaz's key (step 6) identifies the length ascospore group (5–7.5 µm) where *E. kochiana* appears to belong, the length range of its 4.5–6 µm diam ascospores does not overlap with that in the Hawaiian specimen.

Recently, *E. alsophila*, *E. arecae*, *E. comosa*, *E. gliricidae*, and *E. kochiana* were transferred to the re-instated genus *Peroneutypa* Berl. based on ascus morphology (Carmarán et al. 2006). The asci in this genus were described as urn-shaped but with a truncated apex and wider in the middle where ascospores tend to cluster. The apical portion has a thick wall and very small apical ring and lacks any channel. The asci in *Eutypella giffardiani* from Hawaii have a thick-walled apical region that is penetrated by a narrow channel with cytoplasmic strands connecting the apex with the ascus cytoplasm. This kind of ascus is considered to be typical of true species of *Eutypella* (Carmarán et al. 2006).

After the publication of Rappaz's (1987) monograph, several new species of *Eutypella* were described (Agarwal & Gupta 1988; Rajak et al. 1988; Ananthapadmanaban 1989), some of which form ascospores averaging 5–8 µm long (e.g., *E. pongamiae* G.P. Agarwal & S. Gupta). However, as information about the iodine reaction of the ascal apical ring and ostiole size is lacking, it is difficult to make a proper comparison. *Eutypella ceibae* R.C. Rajak et al. has a comparable ascospore length (4–8 µm), but a width of 2.5–6 µm is indicated (a very unusual range, and data on the ascal apical ring in asci and ostiolar size are also wanting).

Three other species from India — “*E. annonae-squamosae*” A. Pande, “*E. colebrookiae-oppositifoliae*” A. Pande, and *E. rozabaghensis* (Srinivas. & P.G. Sathe) A. Pande, which were originally described in *Quaternaria* Tul. & C. Tul. (Srinivasulu & Sathe 1970, Kale & Kale 1972) and later transferred to *Eutypella* either invalidly (as nom. nov.) or as comb. nov. (Pande 2008) — produce ascospores averaging 7–11 µm long and wider than 3 µm. Therefore, the Hawaiian specimen differs from them.

***Thyridaria hawaiiensis* Lar.N. Vassiljeva & J.D. Rogers, sp. nov.**

FIGS. 2A–E

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*Stromata* valsoidea, mollia, cortice erumpentia, disco ectostromatico pulvinato vel hemisphaerico, castaneo, up to 1.5 mm diam. praedita, intus rubiginosa, ligno circa peritheciis nigra. Perithecia globosa, 300–400 µm diam.; ostiolis leniter papillatis, nigris. Asci longe clavati vel cylindrici, sessiles vel short-stipitati, 100–110 × 12–14 µm, juvenilis



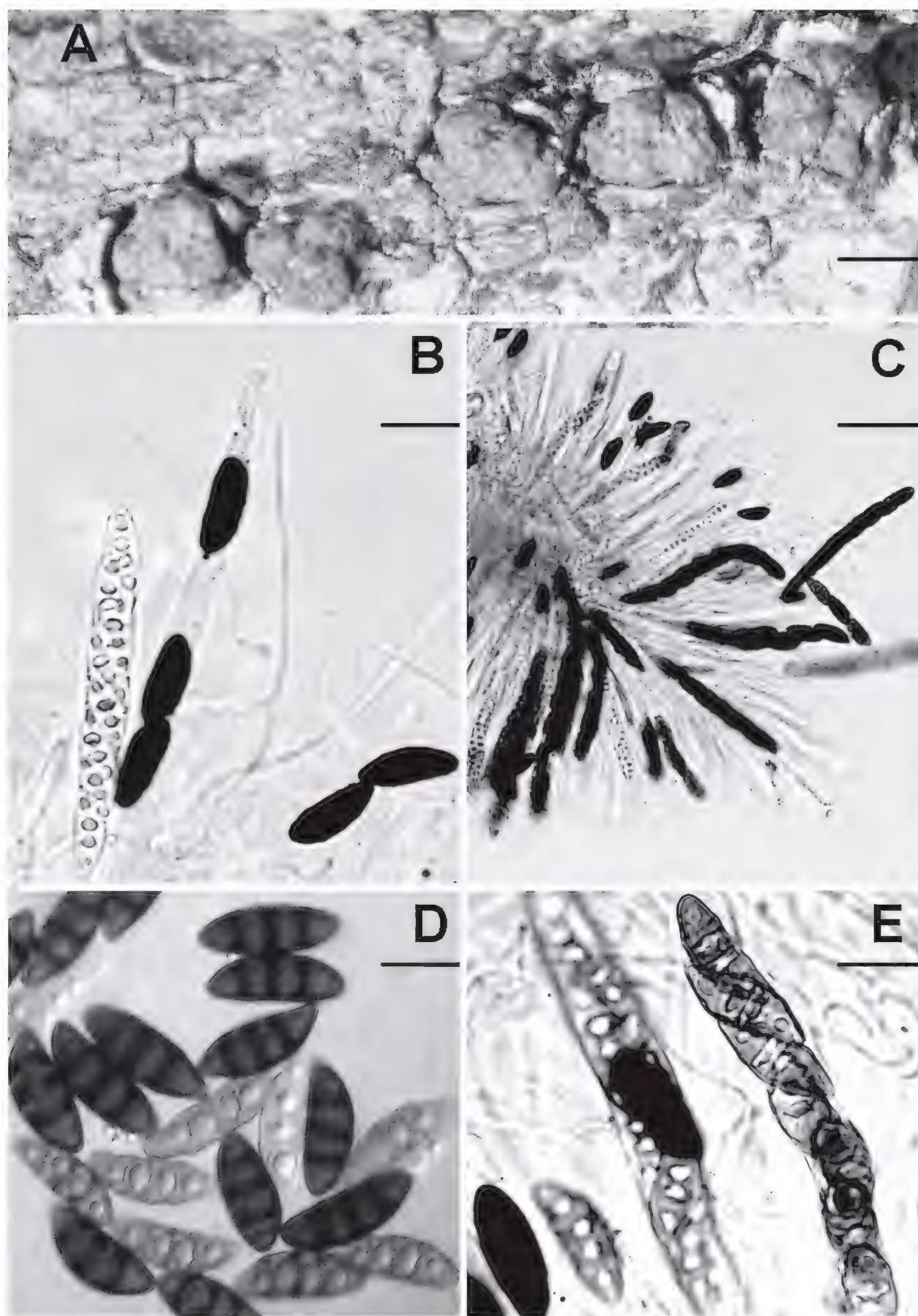


FIG. 2. *Thyridaria hawaiiensis*. A. Stromata. B–C, E. Asci and ascospores. D. Ascospores.  
Scale bars: A = 1 mm; B = 15  $\mu$ m; C = 50  $\mu$ m; D,E = 12  $\mu$ m.

*crasse tunicati; paraphysibus numerosis, hyalinis, longissimis, sinuosis, ca. 1 µm latis. Ascosporae uni- vel biseriatae, ellipsodeae vel fusoideae, rectae, inaequilateralis vel leniter circinatae, 3-septatae, interdum leniter constrictae, pallide- vel atro-fuscae, etiam opacae, levigatae, (19–)20–25(–27) × 6–9 µm.*

**HOLOTYPE:** Hawai'i, Island of Hawaii, Hawaii Volcanoes National Park, Kipuka Puauulu (Bird Park), dead branches of *Hibiscadelphus giffardianus*, 3 November 2005, Jack D. Rogers (BISH). **Isotypes:** VLA; WSP.

**STROMATA** valloid, soft, erumpent from the bark with pulvinate or hemispherical, chestnut (40) ectostromatic 'disc' up to 1.5 mm diam., consisting of confluent and conspicuous tops of perithecial necks with slightly papillate, black ostioles, bay (6) or rust (39) under the surface, black around perithecia deep in the wood. **PERITHECIA** globose, 300–400 µm diam. **ASCI** long clavate to cylindrical, sessile, 100–110 × 12–14 µm, thick-walled when young, surrounded by numerous, hyaline, long, sinuous and anastomosing paraphyses about 1 µm wide. **ASCOSPORES** overlapping uniseriate or irregularly biseriate, ellipsoid-fusoid, straight to inequilateral or slightly curved, 3-septate, not at all or slightly constricted at the septa, light to dark brown, even opaque, smooth, (19–)20–25(–27) × 6–9 µm.

**COMMENTS—** *Thyridaria hawaiiensis* resembles the type of the genus, (*T. incrustans* Sacc.) in ascospore shape, septation, and size, but the latter has a black, carbonaceous disc that is only covered "with a yellow-green pulverulence or pubescence when young" (Wehmeyer 1941). Also, the brown-black entostroma of *T. incrustans* was said to turn reddish in KOH. The stromata of *T. hawaiiensis* are rusty inside, chestnut-colored on the outside, and do not react to KOH.

An earlier name [i.e., *Thyridaria broussonetiae* (Sacc.) Traverso] exists for *T. incrustans* (Barr 1990), which is characterized by ascospores with verruculose walls and apical pores of the perithecia that are bright yellowish to orange pigmented, but the color of the whole stromata is not reported. The apical pore pigmentation is more similar to that found in some *Byssosphaeria* species that were placed together with *Thyridaria* in the *Melanommatales* (Barr 1990). The smooth ascospores in *T. hawaiiensis* distinguishes it from the verruculose spored *T. broussonetiae*.

Another endemic Hawaiian plant, *Acacia koa* A. Gray, was reported to support a separate *Thyridaria* species, namely *T. koae* Petr. characterized by smaller ascospores—14–21 (mostly 18) × 6–9 µm (Petrak 1952).

***Valsonectria macrospora*** J.D. Rogers & Lar.N. Vassiljeva, **sp. nov.**

**FIGS. 3A–D**

MYCOBANK MB 518318

*Stromata cortice erumpentia, pulvinata, 1–2 mm diam, gregaria, lignose ubi siccata vel molliia ubi madefacta, extus initio aurantiaca demum castanea, intus aurantaca sine pigmento in KOH. Perithecia 0.2–0.3 mm diam, ambitus distincta vel indistincta,*



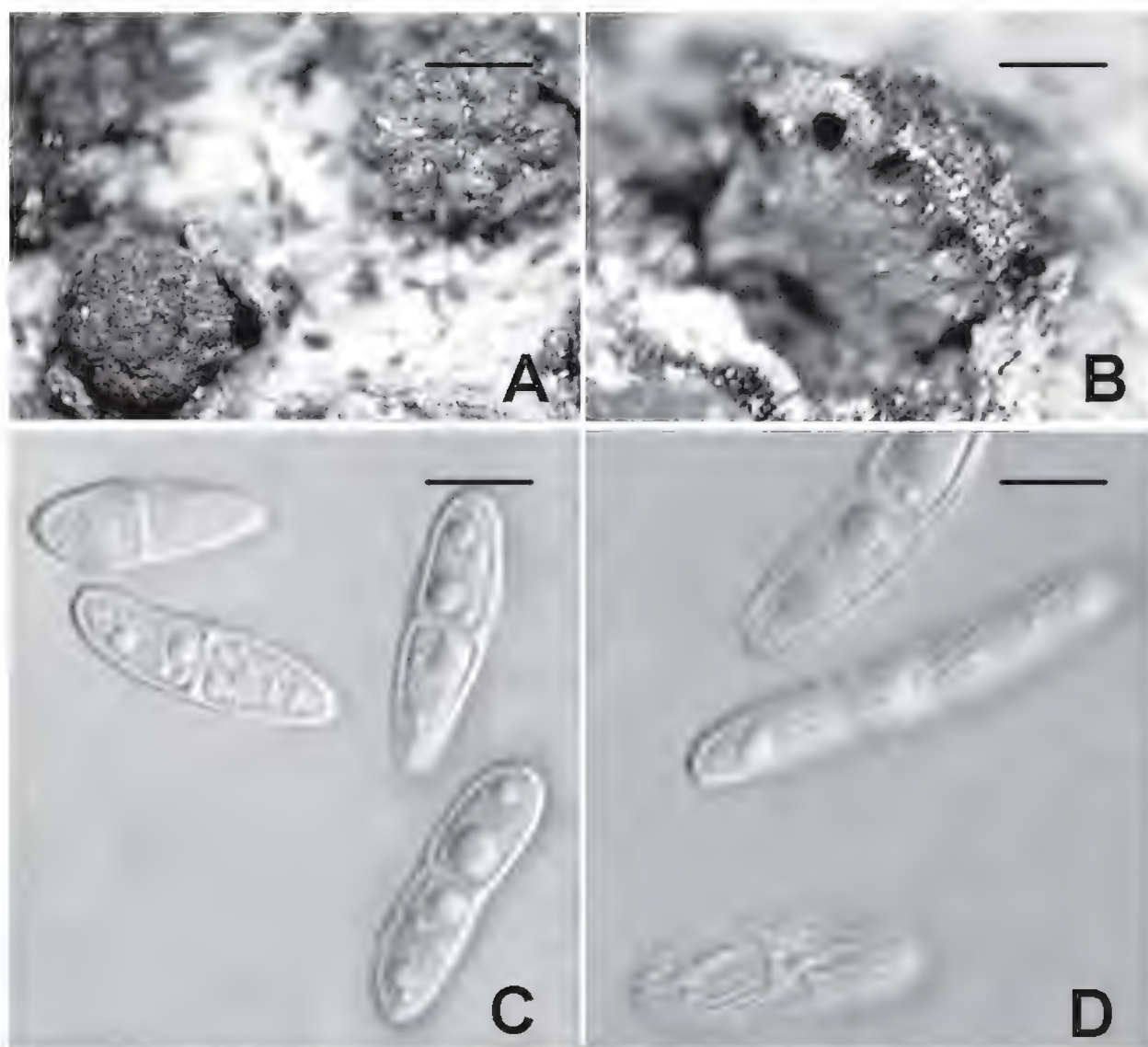


FIG. 3. *Valsonectria macrospora*. A–B. Stromata. C–D. Ascospores.  
Scale bars: A = 1 mm; B = 0.5 mm; C, D = 10  $\mu$ m.

*monosticha*. Ostiola papillata, minuta. Asci octospori, ascosporis irregulariter dispositis, ca. 100  $\mu$ m longitudine tota, ca. 18  $\mu$ m crassi, partibus sporiferis ca. 91  $\mu$ m longitudine, annulo apicali nullo. Asci infrequenter observatis, procliter deliquescentibus. Ascosporae hyalinae, pariter bicellulares, ellipsoideae vel aliquantum inequilaterales, striis longitudinalibus ornatae, 28–31(–46)  $\times$  9–12(–15)  $\mu$ m, sine poro vel rima germinationis. Paraphyses vel pseudoparaphyses apparenter nullae.

**HOLOTYPE:** Hawai'i, Island of Hawaii, Hawaii Volcanoes National Park, Kipuka Puau (Bird Park), dead branches of *Hibiscadelphus giffardianus*, 3 November 2005, Jack D. Rogers (BISH).

**STROMATA** erumpent from bark, pulvinate, 1–2 mm diam, gregarious, woody when dry, soft when wet, surface at first orange (7) becoming chestnut (40), interior orange (7), not releasing a pigment in KOH. **PERITHECIA** 0.2–0.3 mm diam with contours distinct or obscure, monostichous, ostioles papillate, minute. **ASCI** eight-spored, the ascospores jumbled, ca. 100  $\mu$ m total length, 18  $\mu$ m broad, spore-bearing part ca. 91  $\mu$ m long, with apex not bluing in Melzer's iodine reagent. **ASCI** infrequently observed, probably deliquescent. **ASCOSPORES**

hyaline, equally 2-celled, ellipsoid to somewhat inequilateral, ornamented with longitudinal striations,  $28\text{--}31(-46) \times 9\text{--}12(-15) \mu\text{m}$ , without germination pore or slit. Hamathecial elements not seen.

COMMENTS—The material described herein, although abundant, seems overmature. It is included here because the ascospore average is much longer and broader than any other described *Valsonectria* species (Rossman et al. 1999). The striate ornamentation as observed by light microscopy (FIGS. 3D) are, in reality, ribs when seen by SEM. Asci, which are not frequently encountered, appear to be unitunicate and are probably deliquescent. Hamathecial elements appear to be absent, but this might be due to the apparent overmaturity of the material. Our late colleague, Margaret Barr, examined our material and believed it to be a *Valsonectria*. It is noteworthy, however, that several species first described in *Valsonectria* have been transferred to *Valsaria* (Ju et al. 1996; Rossman et al. 1999). It is likewise noteworthy that none of the species of *Valsonectria* recognized by Seifert & Samuels (1997) has a valsoid arrangement of perithecia. Consequently, the name *Valsonectria* actually implies a feature that is not extant.

### Acknowledgments

We thank Don Hemmes, Hilo, for obtaining permission for JDR to collect in Hawaii Volcanoes National Park, for his companionship, for identifying hosts, and for other aid that furthered these studies. We are also grateful to Dean A. Glawe, Washington State University, and Steven L. Stephenson, University of Arkansas, for their helpful comments to this paper.

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## MYCOTAXON

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**New names in the genus *Marasmius***

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**Abstract** — Illegitimate later homonyms for six well documented species in the genus *Marasmius* are used in recent literature. Consequently, new names are proposed: *M. asiaticus* (= *M. distantifolius* Y.S. Tan & Desjardin), *M. canalipes* (= *M. sulcatipes* Pat.), *M. leelavathyi* (= *M. parvulus* Manim. & Leelav.), *M. lilacinitinctus* [= *M. lilacinus* (Coker & Beardslee) Singer], *M. masseei* (= *M. aratus* Masee), and *M. neotropicus* (= *M. asemus* Singer).

**Key words** — *Agaricales*, *Basidiomycota*, *Marasmiaceae*, nomenclature

**Introduction**

In recent literature dealing with the genus *Marasmius* Fr., we have noticed several later homonyms of validly published names. Six later homonyms are used for well-documented species: *Marasmius aratus* (Masee 1914), *M. asemus* (Singer 1989), *M. distantifolius* (Tan et al. 2009), *M. lilacinus* (Singer 1951), *M. parvulus* (Manimohan & Leelavathy 1987), and *M. sulcatipes* (Patouillard 1924). These names are illegitimate according to Art. 53.1 of the International Code of Botanical Nomenclature (McNeill et al. 2006). Therefore, we propose new names for these six species.

**Taxonomy**

***Marasmius asiaticus*** Mešić & Tkalčec, nom. nov.

MYCOBANK MB 518123

= *Marasmius distantifolius* Y.S. Tan & Desjardin, Fungal Diversity  
37: 95, 2009, nom. illeg., non (Murrill) Murrill 1915.

ETYMOLOGY: The species is named after the continent on which it was found.

The species belongs to the section *Sicci* Singer. Tan & Desjardin (Tan et al. 2009) described this species based on one collection from Peninsular Malaysia.



***Marasmius canalipes*** Tkalčec & Mešić, nom. nov.

MYCOBANK MB 516949

= *Marasmius sulcatipes* Pat., Bull. Mus. Natl. Hist. Nat. 30:  
526, 1924, nom. illeg., non Murrill 1915.

ETYMOLOGY: The species is named for the striate surface of its stipe.

The species belongs to the section *Globulares* Kühner. Since it was originally described on the basis of a single collection from Madagascar by Patouillard (1924), it has never been found again. Antonín & Buyck (2006) made an analysis of the holotype, redescribed the micromorphological characters, and compared it with similar species.

***Marasmius leelavathyi*** Manim., Tkalčec & Mešić, nom. nov.

MYCOBANK MB 518137

= *Marasmius parvulus* Manim. & Leelav., Trans. Brit. Mycol. Soc. 88(3):  
422, 1987, nom. illeg., non Berk. & M.A. Curtis 1860.

ETYMOLOGY: Dedicated to Prof. K. M. Leelavathy, Indian mycologist.

The species belongs to the section *Neosessiles* Singer. Manimohan & Leelavathy (1987) described it based on only one collection from India. Thereafter, it has not been found again (P. Manimohan, pers. comm.).

***Marasmius lilacinitinctus*** Mešić & Tkalčec, nom. nov.

MYCOBANK MB 518141

= *Collybia lilacina* Coker & Beardslee, J. Elisha Mitchell Sci. Soc. 37(1): 104, 1921.  
= *Gymnopus lilacinus* (Coker & Beardslee) Murrill, Mycologia 30(4): 367, 1938.  
= *Marasmius lilacinus* (Coker & Beardslee) Singer, Lilloa 22:  
326, 1951 ["1949"], nom. illeg., non Henn. 1896.

ETYMOLOGY: The species is named for its lilac tones on basidiomata.

Coker & Beardslee (1921) described this species in the genus *Collybia* (Fr.) Staude. Singer (1951) transferred it to the genus *Marasmius* where it is classified in section *Globulares*. Halling (1983) redescribed the species and designated a lectotype. It is distributed in the southeastern part of the USA from North Carolina to Florida (Halling 1983).

***Marasmius masseei*** Tkalčec & Mešić, nom. nov.

MYCOBANK MB 518138

= *Marasmius aratus* Massee, Bull. Misc. Inform. Kew 1914:  
358, 1914, nom. illeg., non W.G. Sm. 1873.

ETYMOLOGY: Dedicated to G. E. Massee, British mycologist.

The species belongs to the section *Sicci*. It was described from Singapore by Massee (1914) and has also been found in Peninsular Malaysia (Tan et al. 2009). For descriptions and comments on similar species, see Corner (1996) and Tan et al. (2009).

***Marasmius neotropicus* Mešić & Tkalčec, nom. nov.**

MYCOBANK MB 518140

≡ *Marasmius asemus* Singer, Fieldiana, Bot., 21: 60, 1989,  
nom. illeg., non (Fr. : Fr.) P. Karst. 1889.

ETYMOLOGY: The species is named after the Neotropical region where it was originally found.

The species belongs to the section *Sicci*. Singer (1989) described it based on two collections from the same locality in Brazil. There are no other records of the species in the literature.

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## MYCOTAXON

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**Macrofungal diversity of Ilgaz Mountain National Park  
and its environs (Turkey)**ILGAZ AKATA<sup>\*1</sup>, BARBAROS ÇETİN<sup>1</sup>, MUSTAFA IŞILOĞLU<sup>2</sup><sup>\*</sup>*fungus@hotmail.com.tr*<sup>1</sup>*Ankara University, Science Faculty, Department of Biology  
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**Abstract** — The current research is based on macrofungi collected from Ilgaz Mountain National Park and its environs between 2004 and 2008. As a result of field and laboratory studies, 220 taxa belonging to 59 families were identified. Nineteen taxa belong to *Ascomycota* and 201 to *Basidiomycota*. Three — *Bisporella subpallida*, *Tricholoma bufonium*, *Leucogyrophana pseudomollusca* — represent new records for Turkish mycobiota. The complete list is available on: <http://www.mycotaxon.com/resources/weblists.html>.

**Key words** — biodiversity, mushrooms, taxonomy

**Introduction**

Ilgaz Mountain National Park is located in a transitional zone between Central Anatolia and the North-West Black Sea region within the boundaries of Çankırı and Kastamonu provinces of Turkey. The national park, which covers 1089 hectares and is situated in the A4 grid (see Davis 1965), has a great importance in terms of its flora, wildlife, geographical location, and natural landscape (Kuter 2008). Among the 109 bryophyte and 630 higher plant taxa identified within national park boundaries, 64 taxa are only indigenous to Ilgaz Mountain (Abay & Çetin, 2003) (FIG. 1).

The region is typical of mountain ranges within the preponctic zone of northwest Anatolia. Most of the area is covered with conifer forests, although angiosperm forests also exist at lower elevations. Fir (*Abies nordmanniana* subsp. *bornmuelleriana* (Mattf.) Coode & Cullen) is the dominant species, sometimes forming mixed stands with beech (*Fagus orientalis* Lipsky), Scots pine (*Pinus*

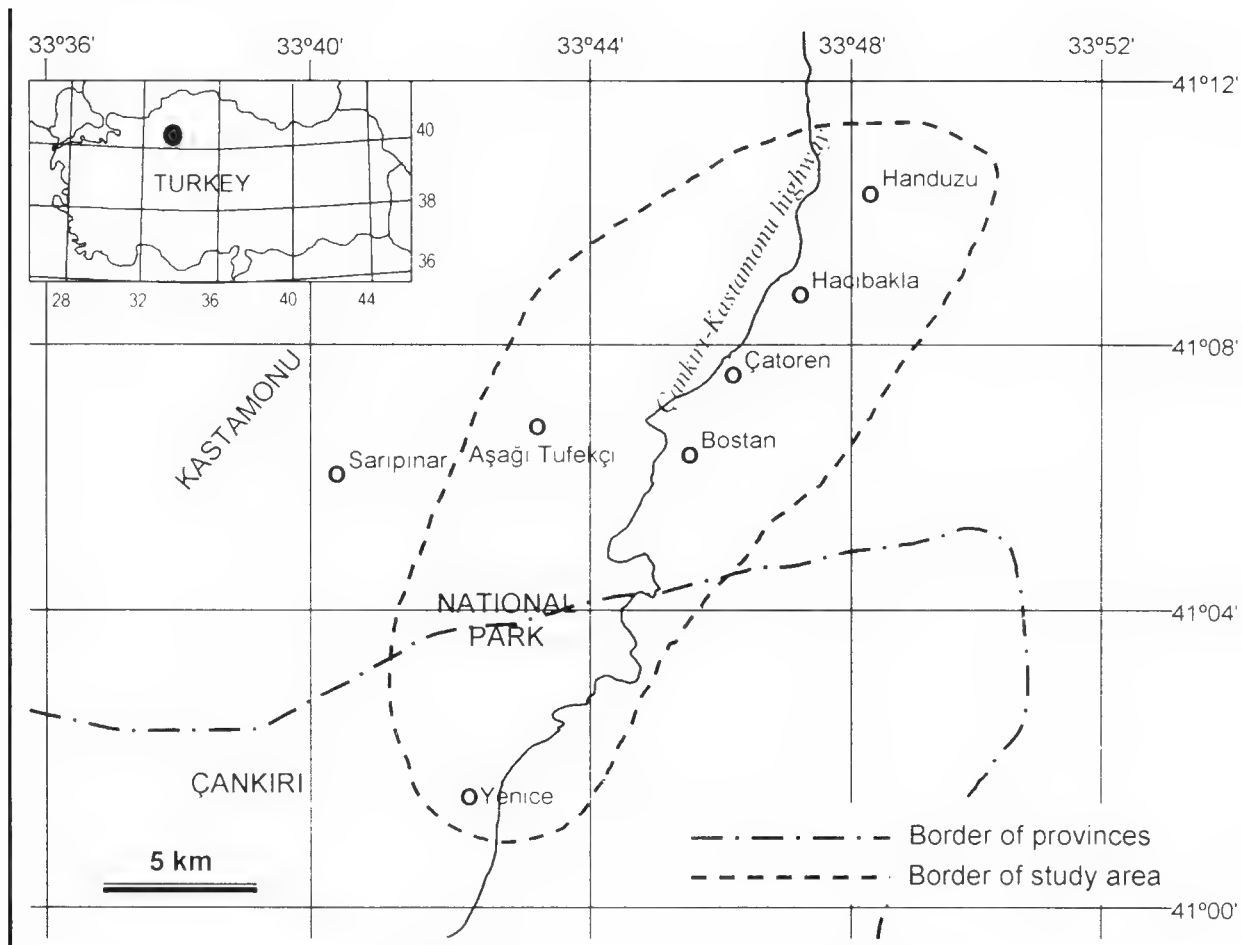


FIGURE 1. Ilgaz Mountain National Park (Turkey).  
Macrofungi collecting area

*sylvestris* L.), and oak (*Quercus petraea* (Matt.) Liebl.). Black pine (*Pinus nigra* J.F. Arnold) and Scots pine are widespread in the western regions of the study area. The southern mountain slopes are influenced by a Mediterranean climate with semi-arid and very cold weather regimes while the northern slopes are under the influence of an oceanic climate (Akman et al. 1983).

Many studies have been conducted on macromycota of Turkey, some of which are still in progress. Sesli & Denchev (2010) cite 1929 macromycete taxa occurring in Turkey based on published research, and Işıloğlu et al. (2010) and Uzun et al. (2010) have contributed additional data. There has not, however, yet been any detailed mycological research devoted to Ilgaz Mountain National Park and its environs.

### Materials and methods

The macrofungi samples of this study were collected from 26 localities in Ilgaz Mountain National Park and its environs between 2004 and 2008. Relevant morphological and ecological characters were recorded for the fungi, which were photographed in their natural habitats. In the herbarium, the fungi were further examined and microscopic characters were measured in Melzer's



reagent, 5% KOH, H<sub>2</sub>O, and H<sub>2</sub>SO<sub>4</sub>. References consulted for identification purposes are provided in the complete annotated species list. All specimens are deposited at the herbarium of Ankara University (ANK).

## Results

As a result of the present study, 220 taxa were identified and named according to the taxonomic conventions of Cannon & Kirk (2007), Kirk et al. (2008), and Index fungorum (www.speciesfungorum.org: accessed 1 January 2010). Taxa are presented in alphabetical order and are listed together with notes on habitat, geographical position, locality, collection date, and accession numbers (A: Akata).

The checklist contains 220 taxa belonging to 124 genera and 59 families. The taxa represent 19 Ascomycota (5 Helotiales, 10 Pezizales, 4 Xylariales) and 201 Basidiomycota (119 Agaricales, 5 Auriculariales, 13 Boletales, 4 Cantharellales, 3 Dacrymycetales, 2 Geastrales, 1 Gloeophyllales, 3 Gomphales, 3 Hymenochaetales, 2 Phallales, 21 Polyporales, 21 Russulales, 3 Thelephorales and 1 Tremellales). Three taxa are new records for Turkey: *Bisporella subpallida* (Rehm) Dennis 1978, *Tricholoma bufonium* (Pers.) Gillet 1874, and *Leucogyrophana pseudomollusca* (Parmasto) Parmasto 1967.

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**First records of *Rhizopogon rocabrunae* and  
*R. pumilionum* (Boletales) from Italy**MIRCA ZOTTI<sup>1\*</sup>, SIMONE DI PIAZZA<sup>1</sup>, ALFREDO VIZZINI<sup>2</sup>

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**Abstract** — The paper reports a macro- and micromorphological investigation on *Rhizopogon rocabrunae*, a very rare hypogeous macrofungus, recently collected in Liguria (Italy) in two different times and locations. These specimens represent the first authentic record of this species from Italy. According to our microscopic analyses, an older Italian collection, formerly identified as *R. rocabrunae*, must be ascribed to *R. pumilionum*, a species previously never reported from Italy. Notes on closely related species are also provided.

**Keywords** — *Abies alba*, *Agaricomycetes*, *Rhizopogonaceae*, *Rhizopogon pannosus*, *Suillineae*

**Introduction**

The genus *Rhizopogon* Fr. encompasses hypogeous ectomycorrhizal fungi primarily associated with members of the *Pinaceae* Lindl. Phylogenetically monophyletic, the genus belongs to the so-called Suilloid radiation or suborder *Suillineae* of the *Boletales* (Grubisha et al. 2001, Binder & Hibbett 2006, Desjardin et al. 2008).

Until now, five species have been reported in Liguria (Northwest Italy), viz. *Rhizopogon luteolus* Fr. 1817, *R. occidentalis* Zeller & C.W. Dodge 1918, *R. rocabrunae*, *R. roseolus* (Corda) Th. Fr. 1909, and *R. villosulus* Zeller 1941. Among the above-mentioned species, *R. rocabrunae* is the least frequent in Liguria, previously reported in Italy on the basis of a single dubious collection (Montecchi & Sarasini 2000) and with few reports from Europe (Martín 1996, Cavet & Lopez 2004). Only two sites are known from Liguria, both located in Ligurian Maritime Alps (Alpi Marittime). One specimen was collected in

Testa d'Alpe forest, in an area dug by wild boars. Testa d'Alpe forest, which extends for 140 hectares from 750 to 1460 m. a.s.l., is the only forest in which the silver fir is considered native in Liguria. The other specimen was observed in an allochthonous silver fir forest, derived from a reforestation.

The paper evaluates the presence of *R. rocabrunae* in Italy based on the study of both the recent Ligurian collections and an older Italian collection (from Lombardia) formerly ascribed to *R. rocabrunae* (Montecchi & Sarasini 2000). Analysis of the old collection was motivated by the fact that Montecchi & Sarasini (2000) illustrated and described specimens with aberrant features for the species and did not mention some microscopic characters that are needed for a correct identification.

### Materials and methods

Macroscopic and microscopic characters were described using a stereo microscope (Leica M 205 C) and a compound microscope (Axioscope, Zeiss), respectively. The description of the features is based on fresh and dry specimens (in the latter case after rehydration in water and lactic acid).

Microscopical observations were made from tissues mounted in distilled water, lactic acid plus acid fuchsin, 5% potassium hydroxide, and Melzer's reagent. For basidiospores and other structures at least 30 individuals were measured. The spore sizes are reported using three numbers corresponding to the minimum, average, and maximum values, respectively. The Qm abbreviation designates the average length to width ratio of the spores in side view.

Colour notations reported in brackets were taken from Kornerup & Wanscher (1978), indicated as "M." in front of a colour code. Identification references were Smith & Zeller (1966), Martín (1996), and Montecchi & Sarasini (2000).

All the Ligurian examined material is deposited and kept at GDOR (Herbarium of the Museo Civico di Storia Naturale Giacomo Doria, Mycologia section, Genova, Italy). Herbarium abbreviations follow Thiers (2010).

As concerns the geo-reference, a Garmin (eTrex Summit) Global Position System (GPS) was set to express the locations in WGS-84 coordinates in decimal degrees. The geographical data were mapped on the Official Map of Italian State (I.G.M.I) using GIS software (MapInfo 7.0). The data were also inserted in a database where all Ligurian macrofungi species are recorded.

### Taxonomy

*Rhizopogon rocabrunae* M.P. Martín, Edic. Espec. Soc. Catalana  
Micol. 5: 95 (1996)

#### Description of the two collections from Liguria

FIG. 1

BASIDIOMATA globose to subglobose, on average 3 cm in diam. PERIDIUM well developed, 0.3–0.6 mm thick, brown with reddish to orange tinges (M. 6 B 8 C 5),

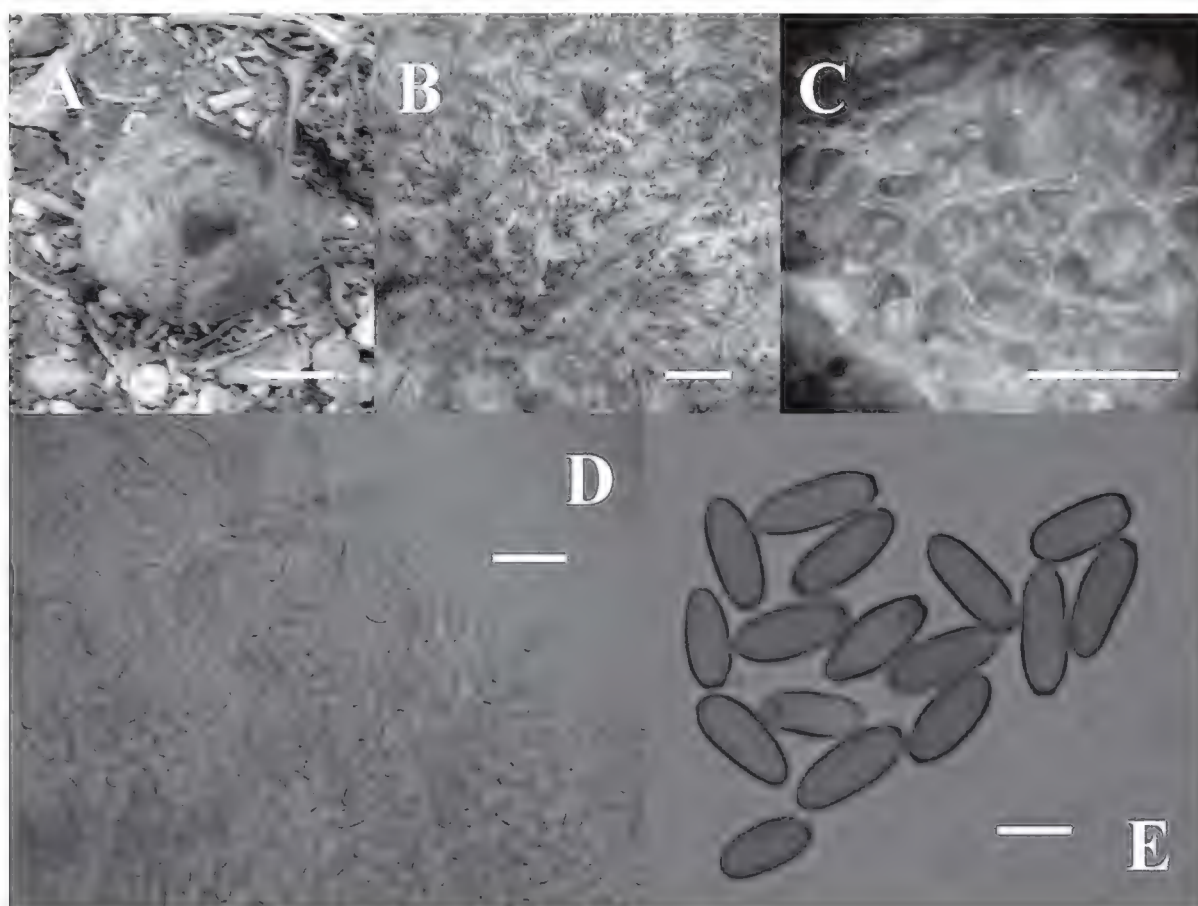


FIGURE 1. *Rhizopogon rocabrunae*.

A. Ripe basidioma. B. Peridium surface. C. Gleba cells. D. E. Basidiospores.

Scale bars: A = 1 cm; B, C = 1 mm; D = 10  $\mu$ m; E = 5  $\mu$ m.

with evident, 0.15–0.35 mm thick squamules, at first orange (M. 6 B 8) then brownish to black. RHIZOMORPHS scarce, gray (M. 1 B, C 1) emanating singly from the base. GLEBA firmly spongy with roundish to elliptical cells, 0.2 to 0.6 mm in diam., yellowish to brownish (M. 5 D 7). TRAMAL PLATES often in part gelatinized 100–200  $\mu$ m thick. SMELL and TASTE indistinct.

PERIDIUM made up of hyaline, septate and thin-walled hyphae, 3–6  $\mu$ m wide, encrusted with brown-orange pigment, with trend mostly parallel to the outer surface; squamules consisting of a more or less parallel arrangement of hyphae, with extracellular orange pigment. BASIDIA cylindrical, 4–10 spored. BASIDIOSPORES (6.6–)7.5(–9)  $\times$  (2.3–)2.9(–3.5)  $\mu$ m,  $Q_m = 2.586$ , elongated, ellipsoidal, smooth, often clearly truncated, transparent yellow to light green (1 A 4, B 4) when ripe, usually pluriguttulate. CLAMP CONNECTIONS absent.

HABITAT – solitary or gregarious under the needle layer of *Abies alba* Mill. Vernal.

MATERIAL EXAMINED: ITALY, Liguria, Foresta Demaniale di Gouta, Testa d'Alpe (IM), 1360 m a.s.l., G.P.S. (wgs 84) long 7.570027° lat 43.945343°, 17/06/2008, leg. M. Zotti, (GDOR 08061701); ITALY, Liguria, Bosco nero, Mendatica (IM) 1350 m. a.s.l., G.P.S. (wgs 84) long 7.733891° lat 44.125178°, 12/06/2008, numerous specimens, leg. G. Baiano (GDOR 08061201).



**Description of the Sarasini collection from Lombardia**

(cited in Montecchi & Sarasini 2000).

BASIDIOMATA globose to subglobose, on average 1–2,5 cm in diam. PERIDIUM color mostly brown (M. 7 F 6 – M. 6 C 4) with reddish tinges (M. 8 C 6). GLEBA with olivaceous tinges (M. 8 C 2). PERIDIUM made up of hyaline hyphae, with trend mostly parallel in the inner layer, while, in the outer layer at the squamules, consisting unordered parallel arrangement of hyphae. BASIDIOSPORES (6.5–)7.93(–9) × (2.2–)2.76(–3) μm, Qm = 2.87, elongated, ellipsoidal, smooth, not always clearly truncated, transparent yellow to light green (1 A 4, B 4).

HABITAT – solitary or gregarious under the needle layer of *Pinus montana* Mill. In summer.

MATERIAL EXAMINED: ITALY, Lombardia, Valdidentro - Cancano (So), 10/08/1987, consisting of four specimens, leg. Aiana, det. Sarasini (AMB 267)

**Discussion**

Martín (1996) originally described *R. rocabrunae* from Spain based on two collections found under *Abies alba*. In the last ten years it has been reported from Italy under *Pinus montana* s.l. (Montecchi & Sarasini 2000) and from France in a *Picea alba*-*Abies alba* wood (Cavet & Lopez 2004).

The two Ligurian collections show features fitting very well the original description of *R. rocabrunae* (Martín 1996). This species is macroscopically characterized by a reddish orange squamulose peridium and microscopically by elongate basidiospores, clearly truncate at the base, and peridial squamules made up of hyphae running parallel to the peridium surface. The squamulose peridium gives the basidiomes a quite distinct and characteristically *Elaphomyces*-like appearance or, as reported by Martín (1996), resembling *Arbutus* berries.

*Rhizopogon rocabrunae* comes very close to *R. pannosus* Zeller & C.W. Dodge 1918, a North American species (Smith & Zeller 1966) reported also from Spain (Martín 1996) and recently from Switzerland (Kathriner & Mühlebach 2008). The latter species differs in having a more verrucose peridium with more irregular squamules made up of interwoven hyphae running perpendicular to the peridium surface, less gelified hyphae of the tramal plates, wider basidiospores (on average 3.4 μm, Qm = 2.3), different isoenzymatic and PCR-RFLP patterns (Martín 1996, Martín & Sánchez 1996, Moser & Peintner 2000), and an association with *Pinus* spp.

According to Moser & Peintner (2000), *R. pumilionum* (Ade) Bataille 1923 from the Austrian *Pinus montana* forests shares with *R. pannosus* the same structure of the peridial squamules, but it is distinguished by its narrower spores (on average 2.9 μm, Qm = 2.6; Moser et al. 1999, Moser & Peintner 2000). Preliminary studies based on 28S rDNA analysis by Jarosch (2001)

indicate that *R. rocabrunae*, *R. pannosus*, and *R. pumilionum* are closely allied but independent species.

The Sarasini collection from Lombardia labelled *R. rocabrunae* (Montecchi & Sarasini 2000) seems quite anomalous due to the ochraceous-coloured peridium and its association with *Pinus montana*. Microscopical analysis revealed that it is referable to *R. pumilionum* based on peridial squamules made up of tufts of ascending hyphae and on spore size. Additionally, the olivaceous tinged gleba and association with *P. montana* are features typical for this species (Moser & Peintner 2000). Therefore, this collection represents the first record of *R. pumilionum* from Italy.

The two Ligurian specimens represent the first authentic report of *R. rocabrunae* from Italy. Adding to the original Spanish and French collections, our records confirm that *Abies alba* seems to be the preferred ectomycorrhizal partner of *R. rocabrunae*, highlighting, as already pointed out in other *Rhizopogon* species as well as in related genera (e.g. *Suillus*), a rather strict, specific association between the mycobiont and the photosynthetic host in the *Suillineae* (Grubisha et al. 2001, 2002).

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Our most sincere thanks are due to María P. Martín (Real Jardín Botánico, Madrid, Spain) and to Giuseppe Venturella (Dipartimento di Scienze Botaniche, Palermo, Italy) for their pre-submission reviews.

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## MYCOTAXON

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**New species of *Dendryphiopsis* and *Stauriella* from Goa, India**J. PRATIBHA<sup>1</sup>, S. RAGHUKUMAR<sup>1</sup> AND D.J. BHAT<sup>2</sup>*jalmipratibha@rediffmail.com**s\_raghukumar@mykotech.com & bhatdj@rediffmail.com*<sup>1</sup>*Myko Tech Pvt. Ltd., Plot no. 12, Mapusa Industrial Estate  
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**Abstract** – Two new species of hyphomycetes isolated from decaying plant litter collected from Goa, India, are described and illustrated. *Dendryphiopsis goanensis*, found on decaying bark of an unidentified tree, is characterized by mostly polytretic, integrated, discrete, terminal, and intercalary conidiogenous cells. *Stauriella indica*, collected from decaying spathe of coconut tree, is characterized by sub-hyaline, spinulate, staurosporous conidia with 15–20 cells.

**Key words** – biodiversity, taxonomy

**Introduction**

During the course of studies on microfungi from forests of Western Ghats in Goa, two hitherto undescribed hyphomycete species, belonging to the genera *Dendryphiopsis* S. Hughes and *Stauriella* Sivichai & E.B.G. Jones, were isolated from fallen and decaying plant litter. Description and illustration of these fungi form the subject matter of this paper.

**Taxonomic descriptions*****Dendryphiopsis goanensis* Pratibha, Raghuk. & Bhat, sp. nov.**

FIGS. 1, 2

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*Ad fungos conidiales, hyphomycetes. Coloniae in substrato naturali dispersae, atrobrunneae vel nigrae; mycelium partim superficiale, partim substrato immersum, ex hyphis laevibus, pallide brunneis, ramosis, septatis, 2–2.5 µm latis, compositum. Coloniae in PDA-cultura, viridi-brunneae, lanatus, reverses nigrae, margin serratus, diam. 2.1 cm aetate 10 dierum. Stroma nullus. Conidiophora macronematica, mononematica, singula vel laxe fasciculata, erecta, recta vel leviter flexuosa, ramosa ad apicem, atrobrunnea, multiseptata, 85–230 × 4–6 µm. Cellulae conidiogenae monotreticae et polytreticae, in conidiophoris incorporatae*

*et discretas, terminales et intercalares, calyciformes, 7.5–13.5 × 4.5–7 μm. Conidia solitaria, cylindrica, utrinque rotundata, atro brunnea, laevia, 3–5-septata, 20–40 × 5–7.5 μm.*

**HOLOTYPE:** On dead and decaying bark of unidentified tree, 13/11/2008, Pratibha J., Mashem, Canacona, Goa, India, Herb. No. HCIO 49724.

Conidial fungi, hyphomycetes. Colonies on natural substrate scattered, dark brown to black. Mycelium partly superficial, partly immersed in the host tissue, composed of smooth, light brown, branched, septate, 2–2.5 μm wide hyphae. Colonies on PDA greenish-brown, wooly, reverse black, margin serrated, attaining a diam. of 2.1 cm in 10 days. Stroma none. Conidiophores macronematous, mononematous, single to loosely fasciculate, erect, straight to slightly flexuous, branched at the apex, dark brown, multiseptate, 85–230 × 4–6 μm. Conidiogenous cells mostly polytretic, sometimes monotretic, integrated, discrete, terminal and intercalary, calyciform, 7.5–13.5 × 4.5–7 μm. Conidia solitary, cylindrical, rounded at both the ends, dark brown, smooth, 3–5-septate, 20–40 × 5–7.5 μm.

NOTES: Hughes (1953) established the genus *Dendryphiopsis* with *D. atra* as type species to accommodate *Dendryphion atrum* Corda. Later Hughes (1958) added two species, *Dendryphiopsis arbuscula* and *D. fascicularis*. Subsequently, two new species have been described in *Dendryphiopsis*, *D. biseptata* (Morgan-Jones et al. 1983), and *D. binsarensis* (Subramanian & Srivastava 1994). Thus, the genus until now has accommodated five species, which are characterized by monotretic, discrete, cylindrical conidiogenous cells and pigmented, thick-walled conidia with two or more transverse septa (TABLE 1). *D. goanensis* differs from earlier described species by having conidiogenous cells that are polytretic, integrated as well as discrete, and terminal as well as intercalary.

TABLE 1: Synopsis of *Dendryphiopsis* spp.

SPECIES	CONIDIOPHORES (μm)	CONIDIOGENOUS CELLS	CONIDIA (μm)
<i>D. arbuscula</i>	240–580 × 10–13	Monotretic, integrated or discrete, terminal, determinate	3–5-septate, 42–64 × 12–14
<i>D. atra</i>	200–400 × 8–11	Monotretic, integrated or discrete, terminal, determinate or percurrent	2–5-septate, 35–65 × 13–20
<i>D. binsarensis</i>	280–520 × 6.5–8	Monotretic, subconical, truncate at apex	4–5-septate, 36–44 × 8–10
<i>D. biseptata</i>	180 long, 8–10 wide	Monotretic, integrated or discrete, cylindrical or narrowly clavate	2-septate, 28–39 × 19–22
<i>D. fascicularis</i>	200–450 × 9–11	Monotretic, integrated or discrete, cylindrical or narrowly clavate	3–8-septate, 48–90 × 5–10
<i>D. goanensis</i>	85–230 × 4–6	Mostly polytretic, sometimes monotretic, terminal or intercalary, integrated or discrete	3–5-septate, 20–40 × 5–7.5



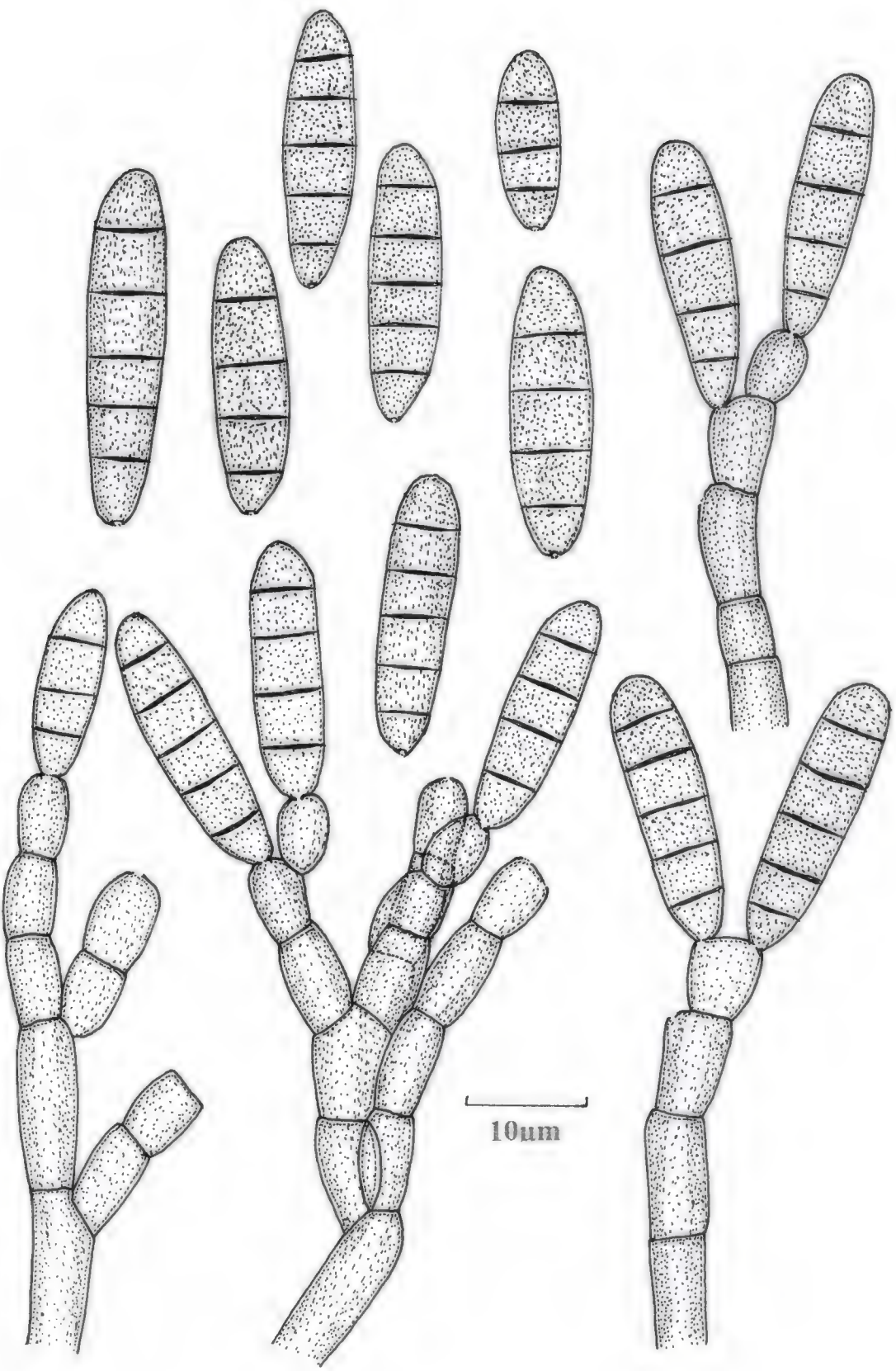


FIG. 1. *Dendryphiopsis goanensis*.  
Conidiophores, conidiogenous cells, and conidia

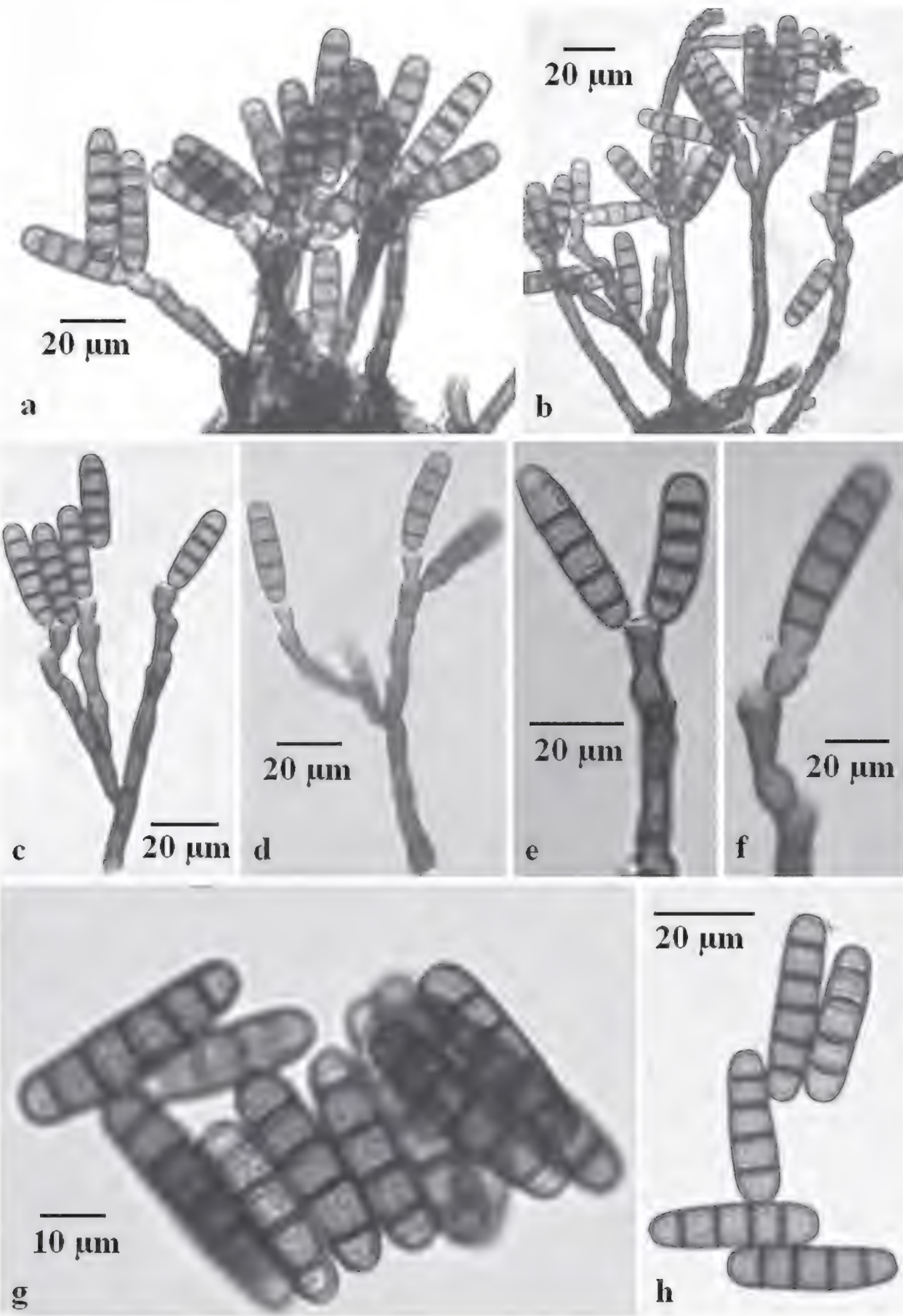


FIG. 2. *Dendryphiopsis goanensis*.  
a–f. conidiophores, conidiogenous cells, and conidia; g–h. conidia

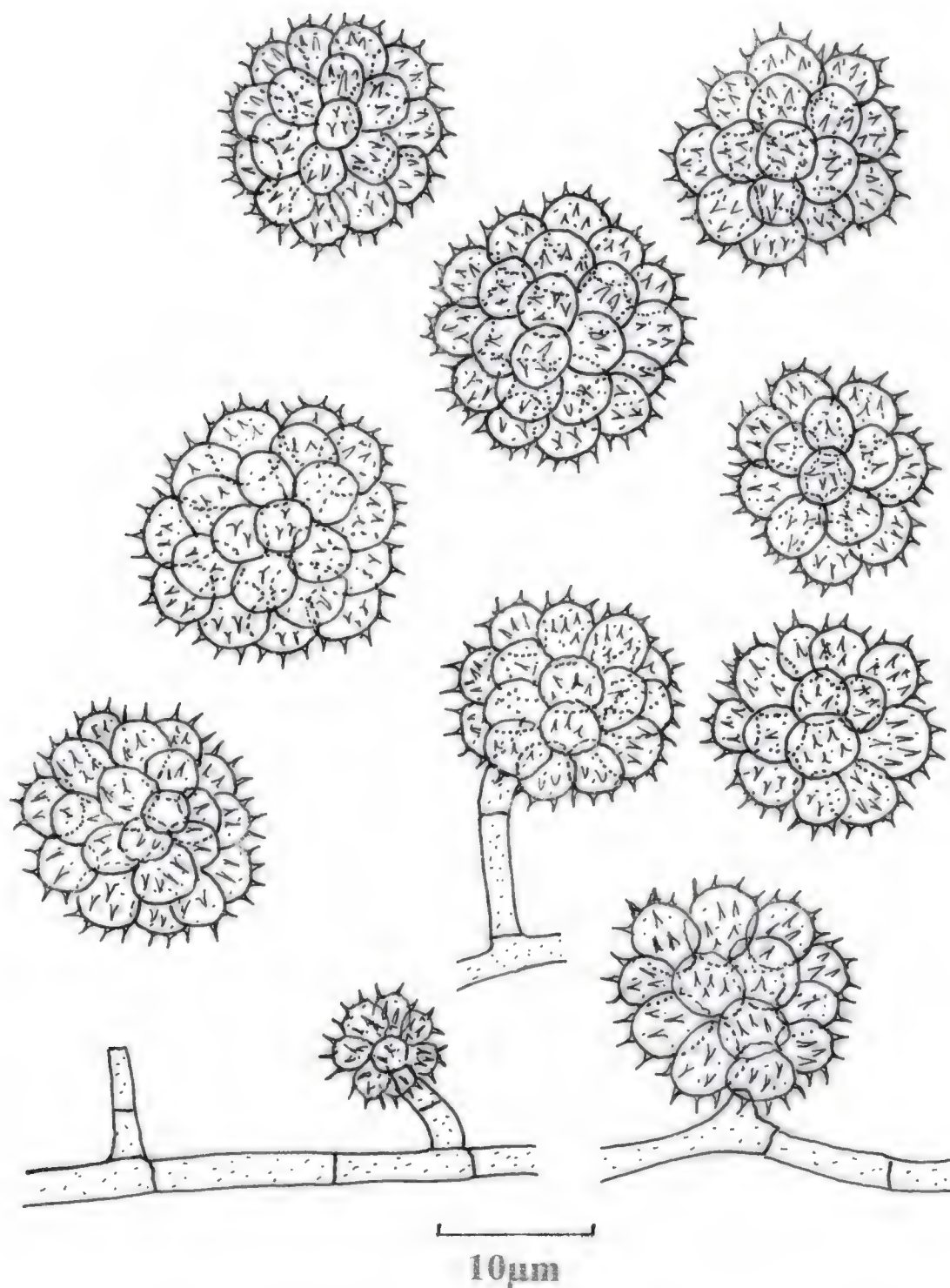


FIG. 3. *Stauriella indica*.  
Conidiophores, conidiogenous cells, and conidia

***Stauriella indica*** Pratibha, Raghuk. & Bhat, sp. nov.

FIGS. 3, 4

MYCOBANK MB 516559

*Ad fungos conidiales, hyphomycetes. Coloniae in substrato naturali effusae, candidae; mycelium partim superficiale, partim substrato immersum, ex hyphis laevibus, hyalinis, ramosis, septatis, 2–3 μm latis, compositum. Stroma nullus. Conidiophora semi-macronematica, mononematica, laevia, hyalina. Cellulae conidiogenae monoblasticae, terminales, integratae, hyalinae, usque ad 10 μm longus, lateraliter orientes. Conidia sicca,*



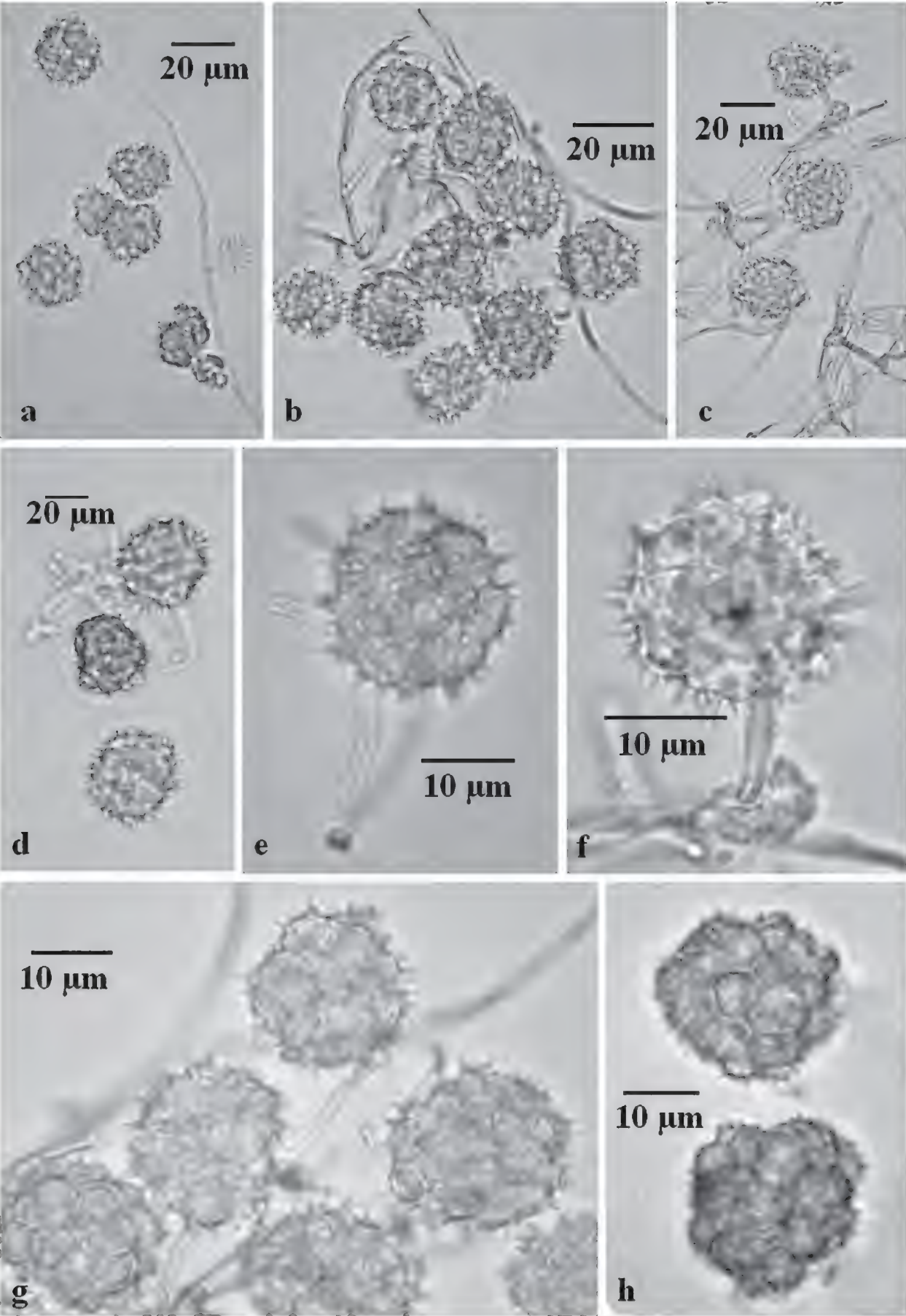


FIG. 4. *Stauriella indica*.  
a–g. conidiophores and conidia; h. conidia

*solitaria*, *hyalina* vel *subhyalina*, 17–23.5  $\mu\text{m}$  diam., ex cellula basali et 15–20 cellulis, cum numerosus spines in omnibus cellulis, conformata.

**HOLOTYPE:** On decaying spathe of *Cocos nucifera*, 17/11/2008, Pratibha J., Mashem, Canacona, Goa, India, Herb. No. HClO 49725.

Conidial fungi, hyphomycetes. Colonies on natural substrate effuse, dull white. Mycelium partly superficial, partly immersed in the host tissue, composed of smooth, hyaline, branched, septate, 2–3  $\mu\text{m}$  wide hyphae. Stroma none. Conidiophores semi-macronematous, mononematous, smooth, hyaline. Conidiogenous cells monoblastic, terminal, integrated, hyaline, up to 10  $\mu\text{m}$  long, arising laterally from hyphae. Conidia dry, solitary, hyaline to sub-hyaline, 17–23.5  $\mu\text{m}$  in diam., comprising 15–20 cells, each with numerous spines on the surface.

NOTES: Sivichai & Jones (2004) established the genus *Stauriella* with *S. aquatica* as type species to accommodate a fungus with hyaline, multicelled, spinulate conidia. The genus was so far monotypic. *S. indica* differs from the type species with conidia comprising 15–20 cells, each with numerous spines and measuring 17–23.5  $\mu\text{m}$  in diam. The conidia in *S. aquatica* are 4–6 celled, each with 2–6 spines and 10–12.5  $\mu\text{m}$  diam.

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## MYCOTAXON

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**A new species of *Graphis* (lichenized Ascomycetes)  
from South Korea**YOGESH JOSHI<sup>1</sup>, ROBERT LÜCKING<sup>2</sup>, YOSHIKAZU YAMAMOTO<sup>3</sup>,  
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**Abstract** — *Graphis flavopalmicola* is described as a new lichenized fungus from Jeju Island (South Korea). It is characterized by smooth, whitish-gray, UV+ pale yellow thallus (lichexanthone), unbranched to irregularly branched lirellae; completely carbonized exciple, and transversely 5–9-septate ascospores. It differs from the closely related *G. palmicola* chiefly in its chemistry; the latter has no substances and is UV–.

**Key words** — biodiversity, *Graphidaceae*, lichens, *Ostropales*, taxonomy

**Introduction**

The lichen genus *Graphis* is characterized by a crustose thallus, rounded to lirellate or rarely pseudostromatic ascomata with carbonized exciples; non-amyloid, functionally unitunicate asci with apical wall thickenings, hyaline, amyloid ascospores with lens shaped lumina, and a trentepohlioid photobiont (Staiger 2002; Lücking 2009). The genus is represented by more than 300 species in the world (Kirk et al. 2008; Lücking et al. 2009). Recent molecular study has confirmed the placement of the genus *Graphis* within family *Graphidaceae* (Mangold et al. 2008).

In South Korea, this genus has so far been investigated to a limited extent with records of only nine species (Kim & Lee 1975; Kim 1976, 1981; Ka et

al. 1997; Park 1982; Hur et al. 2005). During the course of floristic surveys in the extreme southern part of South Korea (Jeju Island), an unknown species of *Graphis* was found growing over bark of *Abies* in open canopy forest. It resembled *G. palmicola* Makhija & Adaw. by having a smooth thallus, a completely carbonized exciple, transversely septate spores, and a similar geographical distribution. In this paper, *G. flavopalmicola* is described as new to science based on this specimen.

### Materials and methods

The specimen for this study was collected on Jeju Island, situated in the extreme southern part of South Korea. The material is deposited in the herbarium of the Korean Lichen Research Institute (KoLRI). Description and photographs of external morphology are based on air-dried material observed under a dissecting stereomicroscope (Nikon SMZ645). Sections were made with a razor blade under the stereomicroscope and mounted in lactophenol cotton blue. Anatomical descriptions are based on these preparations under a compound microscope (Nikon Eclipse E200). Ten measurements per apothecial sections were recorded for ascospore dimensions. Iodine test was performed by using Lugol's solution. The chemistry of the specimens was studied with thin layer chromatography (Culberson 1972; Elix et al. 1987; Orange et al. 2001; White & James 1985) using solvents A and C, and high performance liquid chromatography (Yoshimura et al. 1994).

### New species

*Graphis flavopalmicola* Y. Joshi, Lücking & Hur, sp. nov.

FIG. 1

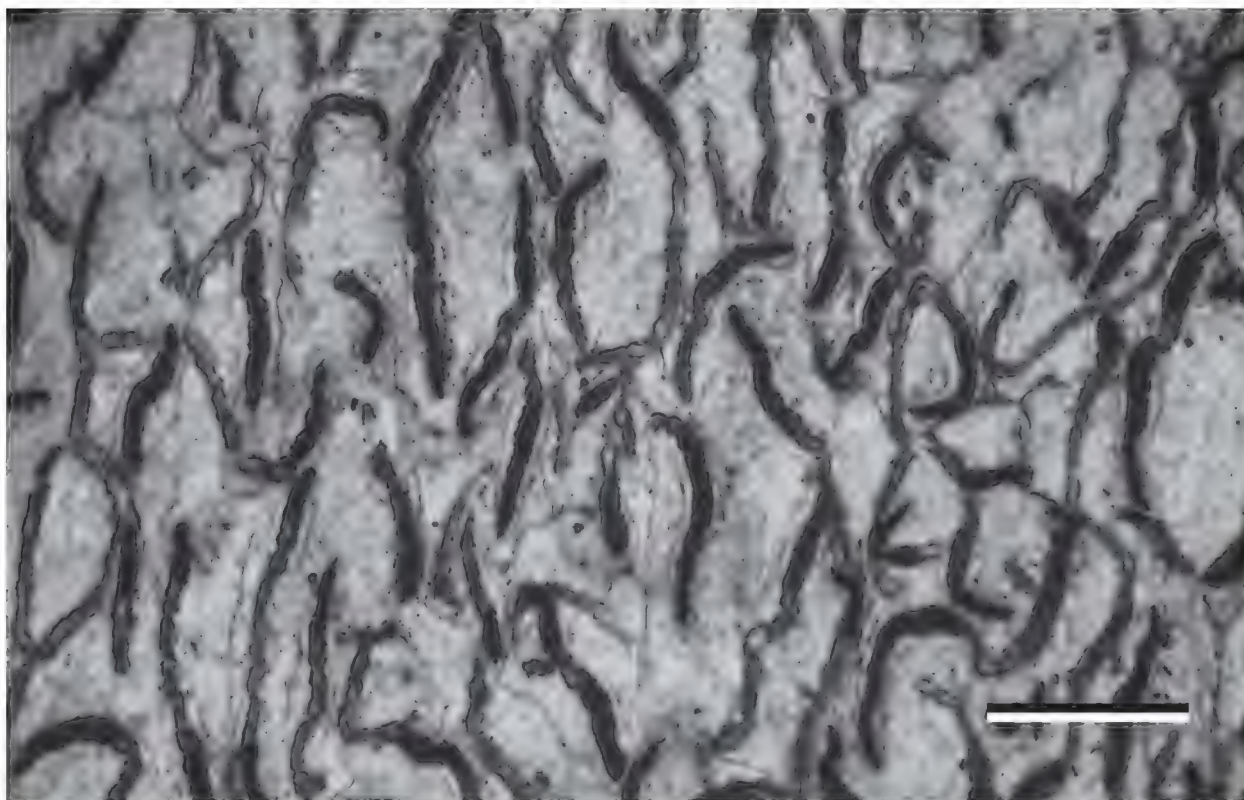
MYCOBANK MB 518424

*Thallus* crustaceus, epiperidermatis, continuus, tenuis, laevigatus vel ob substratum rugulosus, albus vel glauco-cinereus, opacus, UV+ flavescens. Ascomata lirellina, erumpentes, simplicia vel aniso-dichotomiter vel aniso-trichotomiter ramosa, flexuosa, usque ad 6 mm longa et 0.2 mm lata, apicibus acutus vel obtusus. Discus foramen, nigrus, epruinosis. Labia convergentia. Excipulum in toto carbonaceum. Hymenium non-inspersum. Paraphyses filiformes, simplices, densae, ad apicem modica clavatae et fulvescentes. Asci (4–)8-spori, ascosporae oblong-fusiformes, rectae, ad apicem rotundatae vel angustato-rotundatae, incolores, I+ caeruleo-violaceae, transversalibus 6–10 loculares, 20–25 µm longae et 4–7 µm latae.

TYPE — SOUTH KOREA, Jeju Island, Mt. Halla, N33°21'99.6", E126°32'15.1", alt. 1714 m, on *Abies* bark, 21 April 2009, Jae-Seoun Hur 090149 (HOLOTYPE- KoLRI, ISOTYPE-KH).

ETYMOLOGY — The species epithet refers to the UV+ yellow thallus and its resemblance to *G. palmicola*.

DESCRIPTION — THALLUS crustose, epiperidermal, continuous, smooth to ± rugulose, 75–100 µm thick, with hyaline crystals scattered in the thallus, but



Habit of *Graphis flavopalmicola* (holotype). Scale = 4 mm.

mainly in clusters near the exciple; surface white or ash-gray, opaque. SOREDIA absent. PROTHALLUS absent.

APOTHECIA much crowded, lirelliform, erumpent, unbranched to rarely anisotomic dichotomously or trichotomously branched, straight to  $\pm$  flexuose, 1–6 mm long and 0.1–0.2 mm wide, terminally acute to obtuse. DISC exposed, black, epruinose (*handelii*-morph according to Lücking 2009). THALLINE MARGIN basal to lateral, but reaching the apices in some lirellae. LABIA entire, convergent. EXCIPIE basally closed, completely carbonized, dirty brown in thin sections, thalline margin with hyaline crystals. EPITHECIUM indistinct, brownish, 5–7.5  $\mu\text{m}$ . HYMENIUM hyaline, not inspersed, I–, 75–140  $\mu\text{m}$  high. PARAPHYSES hyaline, filiform, unbranched, dense, 1–1.5  $\mu\text{m}$  thick, moderately clavate and yellowish brown at apices. ASCOSPORES (4–)8 per ascus, hyaline, transversely 5–9 septate, oblong-fusiform, straight, rounded to narrowly rounded at the apices, (19–)20–25(–27)  $\times$  4–7  $\mu\text{m}$ .

CHEMISTRY — Spot test reactions: thallus K– or yellowish-brown, C–, KC–, P–, UV+ pale yellow. TLC: lichexanthone. HPLC: unknown products at Rt 2.544 and 2.968.

ECOLOGY AND DISTRIBUTION — The species is so far known from the type locality and was found growing over the bark of *Abies koreana* at an elevation of 1714 m. The subalpine forest is mainly composed of *Abies koreana* community.

REMARKS — *Graphis flavopalmicola* is characterized by smooth to rugulose, whitish-gray, UV+ pale yellow thallus, an exposed, blackish disc, entire labia, completely carbonized exciple, and small, transversely septate ascospores. In morphology of the ascomata and general appearance, the new species is most likely to be confused with *G. palmicola*, *G. assimilis* Nyl., and *G. stipitata* A.W. Archer. *Graphis palmicola* differs in having an UV– thallus. *Graphis assimilis* has larger ascospores [23–40(–54)  $\mu\text{m}$ ] and produces norstictic acid (without lichexanthone), while *G. stipitata* differs in having a laterally carbonized exciple, slightly smaller ascospores (15–20  $\mu\text{m}$  long), and the presence of norstictic acid in addition to lichexanthone.

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## MYCOTAXON

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**Elucidating the taxonomic rank of  
*Cladonia subulata* versus *C. rei* (Cladoniaceae)**RAQUEL PINO-BODAS<sup>1\*</sup>, ANA R. BURGAZ<sup>1</sup> & MARÍA P. MARTÍN<sup>2</sup>

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**Abstract** — *Cladonia subulata* and *C. rei* are two lichen species apparently closely related from a morphological viewpoint. Since both species also show a high morphological variability, it has been difficult to establish the limit between them, and their taxonomic classification has often been questioned. Nevertheless, they have different lichen substance contents. The present paper aims to clarify the taxonomy of *C. subulata* and *C. rei*. Their morphological, chemical, and anatomical variation is examined and correlated with the molecular data of three gene regions (ITS rDNA, *rpb2* and *ef1α*). The results of the analyses reveal two strongly supported monophyletic clades, correlated with the two taxa. We conclude that *C. subulata* and *C. rei* should be maintained as two different species.

**Key Words** — Ascomycota, secondary chemistry, sibling species, species delimitation

**Introduction**

The lichens *Cladonia subulata* (L.) F.H. Wigg. and *Cladonia rei* Schaer. can be difficult to distinguish and therefore their taxonomic distinction has recently been questioned, particularly by Spier & Aptroot (2007). Traditionally, they have been regarded as two distinct species in spite of their great morphological similarity. *Cladonia subulata* is even the nomenclatural type species of the large genus *Cladonia* (Ahti 2000). The secondary metabolites, the presence of corticated areas at the base of podetia and the farinose or granular soredia are the main characters used to distinguish those species (Suominen & Ahti 1966, Wirth 1995, Brodo et al. 2001, James 2009).

Paus et al. (1993), who conducted an exhaustive revision of the morphological characters used to differentiate these species, concluded that none of them were sufficient to distinguish the two taxa. Nevertheless, they were attributed

TABLE 1. Specimens included in molecular study and GenBank accession numbers.

Taxon	Code	Chemical	UV	FeCl <sub>3</sub>	Collection	ITS	<i>rpb2</i>	<i>eflα</i>
<i>C. rei</i>	2REI	HSEK	+	+	Canada, Ontario, S L58841	FN868580	HM243200	HM243185
<i>C. rei</i>	3REI	HSEK	+	+	Sweden, Gästrikland, S F52894	FN868581	HM243201	HM243186
<i>C. rei</i>	4REI	HSEK	+	+	Norway, Oslo, BG L86605	FN868582	HM243202	HM243187
<i>C. rei</i>	5REI	HSEK	+	+	Canada, Newfoundland, BG L86394	FN868583	HM243203	HM243188
<i>C. rei</i>	6REI	HSEK	+	+	USA, Minnesota, S F53070	FN868584	HM243204	HM243189
<i>C. rei</i>	7REI	FUM, HSEK	+	+	Spain, Gerona, MACB 92216	FN868585	HM243205	HM243190
<i>C. rei</i>	8REI	FUM, HSEK	+	+	Spain, Barcelona, MACB 100473	FN868586	HM243206	HM243191
<i>C. rei</i>	11REI	HSEK	+	+	Slovakia, Trenčín, BRA 10005	FN868591	-	HM243192
<i>C. rei</i>	12REI	HSEK	-	+	Czech Republic, Central Bohemia, BRA 10044	FN868592	-	-
<i>C. rei</i>	15REI	FUM, HSEK	+	+	Netherlands, Utrecht, Aptroot 68588	FN868590	HM243207	HM243193
<i>C. rei</i>	16REI	HSEK	+	+	Japan, Akita, UPS L170710	FN868593	-	-
<i>C. rei</i>	17REI	FUM, HSEK	+	+	Czech Republic, Karlovy Vary, J. Vondrák 7024	FN868587	HM243208	HM243194
<i>C. rei</i>	18REI	FUM, HSEK	+	+	Czech Republic, South Bohemia, J. Vondrák 7006	FN868588	HM243209	HM243195
<i>C. rei</i>	19REI	FUM, HSEK	+	+	Czech Republic, Karlovy Vary, J. Vondrák 7026	FN868589	-	HM243196
<i>C. subulata</i>	1SUBU	FUM	-	-	Spain, Asturias, MACB 93151	FN868566	HM243210	HM243174
<i>C. subulata</i>	2SUBU	FUM	-	-	Spain, Ávila, MACB 93837	FN868567	HM243211	HM243175
<i>C. subulata</i>	3SUBU	FUM	-	-	Sweden, Gästrikland, S F52879	FN868568	HM243212	HM243176
<i>C. subulata</i>	4SUBU	FUM	-	-	Sweden, Halland, S F90966	FN868569	HM243213	HM243177
<i>C. subulata</i>	5SUBU	FUM	-	-	Spain, Burgos, MACB 97275	FN868570	HM243214	HM243178
<i>C. subulata</i>	6SUBU	FUM	-	-	Spain, Palencia, MACB 95159	FN868577	-	-
<i>C. subulata</i>	7SUBU	FUM	-	-	Spain, La Rioja, MACB 96350	FN868571	HM243215	HM243179
<i>C. subulata</i>	8SUBU	FUM	-	-	Portugal, Trás-os-Montes, MACB 93692	FN868572	HM243216	HM243180
<i>C. subulata</i>	9SUBU	FUM	-	-	Chile, Navarino Island, MACB 92216	FN868578	-	-
<i>C. subulata</i>	12SUBU	FUM	-	-	Slovakia, Moravia, BRA 10048	-	-	HM243181
<i>C. subulata</i>	13SUBU	FUM	-	-	Netherlands, Utrecht, L Spier	FN868573	HM243217	-
<i>C. subulata</i>	15SUBU	FUM	-	-	France, Midi-Pyrénées, L 75293	FN868579	-	-
<i>C. subulata</i>	16SUBU	FUM	-	-	Czech Republic, Central Bohemia, J. Vondrák 6983	FN868574	-	HM243182
<i>C. subulata</i>	18SUBU	FUM	-	-	Denmark, Zealand, J. Vondrák 6967	FN868575	-	HM243183
<i>C. subulata</i>	19SUBU	FUM	-	-	Austria, Upper Austria, FB	FN868576	HM243218	HM243184
<i>C. glauca</i>	1GLAU	SQUA			Spain, Segovia, MACB 96751	FN868594	HM243219	HM243197
<i>C. glauca</i>	3GLAU	BAR, THAM			Spain, Alava, MACB 96090	FN868595	HM243220	HM243198
<i>C. cenotea</i>	1CENO	SQUA			Denmark, Hovedstaden, J. Vondrák 6965	FN868596	HM243221	HM243199

FUM= fumarprotocetraric acid, HSEK= homosekikaic acid, SQUA= squamatic acid, BAR= barbatric acid, THAM = thamnolic acid.

a species rank based on their different habitat preferences. Spier & Aptroot (2007), on the contrary, concluded that as there are not enough characters to maintain the two taxa as independent they represent chemotypes of a single species. Syrek & Kukwa (2008) and James (2009), who have not accepted this viewpoint, retain *C. subulata* and *C. rei* as independent species.

The aim of this study is to resolve the complex *C. subulata*-*C. rei* and attempt to elucidate whether the complex represents two species or chemotypes of the one and the same species. To this end, three gene regions ITS rDNA, *rpb2* and *eflα* have been analyzed in combination with morphological and anatomical characters. Recent studies using DNA sequence data have clarified relationships in several lichen species with high morphological similarities (Argüello et al. 2007, Ohmura & Kanda 2004, Amtoft et al. 2008).

## Material & methods

### Lichen material

A total of 241 specimens of *Cladonia subulata* and 60 of *C. rei* were studied. The samples selected for molecular and morphological study were chosen from several places within the geographical range of these species and are listed in TABLE 1. Some morphologically similar species, such as *C. glauca* Flörke and *C. cenotea* (Ach.) Schaer., were included (Suominen & Ahti 1966, Nourish 1977, Paus 1997, James 2009). *Cladonia cariosa* (Ach.) Spreng. was used as an outgroup because it was basal in the clade where *C. subulata* and *C. rei* were included by Stenroos et al. (2002) in their phylogenetic trees.

### Morphological and chemical data

The samples were identified on the basis of morphology and secondary metabolites. The presence/absence of cortex at the base of podetia, presence/absence of squamules, branching type of podetia (type I: branched antler-like; type II: unbranched or forked at the apex), and cup shape of the podetia were studied macroscopically with a stereomicroscope, and the soredial size was measured under the light microscope. Microscopic measurements of the podetial wall thickness were carried out on sections cut with a freezing microtome. Iodine reactions were tested using Lugol's solution after pre-treatment with 10% KOH. In addition, transverse and lengthwise sections at the base of the podetia were made and stained with lactophenol blue solution. The stereome surface was observed by Scanning Electron Microscopy (SEM) in longitudinal sections of the podetia. Statistical analyses were done by STATGRAPHICS 5.1 computer program. The continuous characters normality and homogeneous variance were subject to analysis of variance (ANOVA) in association with the resulting clades of the phylogenetical analyses. Continuous characters that did not fulfill the normality and homogeneous variance were analyzed by Kruskal-Wallis test. The Kolmogorov-Smirnov test was used to check normality and Levene statistic to check the homogeneous variance. Binary characters were subjected to a test of contingency tables based on  $\chi^2$ -statistic test.

Chemical composition was checked by thin layer chromatography (TLC) according to the standardized procedures of White & James (1985), with solvent systems A and



B. Moreover, 60 samples were visualized under UV light (TABLE 1), and  $\text{FeCl}_3$  reaction (alcoholic dissolution to 10%) was checked on 188 specimens (TABLE 1).

### DNA extraction and PCR

Total DNA was extracted using DNeasy Plant Mini Kit (Quiagen) following the manufacturer's instructions. The DNA was dissolved in 200  $\mu\text{l}$  of buffer included in the kit. Three genetic regions were selected: ITS rDNA, *rpb2* partial gene, and *ef1 $\alpha$*  partial gene. The primers used to amplify the nuclear ITS rDNA were ITS1F (Gardes & Bruns 1993) and ITS4 (White et al. 1990), alternatively 1780-5'F/LSU0012 (Piercey-Normore & DePriest 2001) or ITSclld /ITSclr (Pino-Bodas unpubl. data). The *rpb2* partial gene was amplified using nested PCR. The first PCR was performed with the primer pair RPB2-5F/ RPB2-7cR (Liu et al. 1999); 1  $\mu\text{l}$  of the first amplification served as DNA template for a second reaction using the primers RPB2dRaq (5' GCTGCTAAGTCTACCAT 3') /RPB2rRaq (5' ATCATGCTTGGAATCTC 3') newly designed in this study. The primers used to amplify *ef1 $\alpha$*  partial gene were CLEF-3F/CLEF-3R (Yahr et al. 2006). The amplification program for ITS rDNA was: initial denaturation at 94 °C for 5 min; 5 cycles of 94 °C for 30 s, 54 °C for 30 s and 72 °C for 1 min; and 33 cycles of 94 °C for 30 s, 48 °C for 30 s and 72 °C for 1 min; with a final extension at 72 °C for 10 min. The amplification program for *rpb2* was: initial denaturation at 94 °C for 5 min; 40 cycles of 95 °C for 30 s, 52 °C for 30 s and 72 °C for 2 min; with a final extension at 72 °C for 10 min. The amplification program for *ef1 $\alpha$*  was: initial denaturation at 94 °C for 5 min; 35 cycles of 95 °C for 30s, 55 °C for 30s and 72 °C for 1 min; with a final extension at 72 °C for 10 min. PCRs were carried out with Ready-to-Go-PCR Beads (GE Healthcare Life Sciences, UK). Amplifications were prepared for a 25  $\mu\text{l}$  final volume. PCR was performed using the MJ Reseach-PTC-200 thermocycler (Massachusetts). The PCR products were purified using the QIAquick Kit (QIAGEN, Valencia, California, USA).

### DNA sequencing

The primers for sequencing reactions were those used in PCR amplification. The sequencing reactions were done at the Secugen S. L. (CIB, Madrid, Spain) or Macrogen (Korea) sequencing service ([www.macrogen.com](http://www.macrogen.com)). Sequencher™ program (Gene Codes Corporation, Inc, Ann Arbor, Michigan, USA) was used to assemble the consensus sequence from the two strands of each isolate.

### Sequence alignments and data analysis

The sequences were manually aligned with SE-AL v2.0a11 Carbon (Rambaut 1996) with each region aligned separately. The transitions and transversions were considered for aligning the sequences. The ambiguous positions were removed.

After each gene region was separately analyzed, a matrix combining the three studied gene regions was constructed in which we included only taxa for which sequences of all three gene regions were available. Both individual regions and the combined matrix were analyzed using Maximum Parsimony (MP) and Bayesian Analysis. MP analyses were conducted with PAUP\* version 4.0b10 (Swofford 2002) using heuristic search with 500 replicates and TBR Branch-swapping option. Bootstrap analyses were performed with 10.000 replicates, using the fast-step option. MrModeltest (Nylander 2004) was used for selecting the best evolution model (TABLE 2) for each region. Bayesian analyses were

carried out by MrBayes 3.1 (Huelsenbeck & Ronquist 2001). The posterior probabilities were approximated by sampling trees using Markov Chain Monte Carlo (MCMC). The posterior probabilities of each branch were calculated by counting the frequency of trees visited during MCMC analysis. Model parameters were estimated in each analysis for 2,000,000 generations sampled in 12 simultaneous chains and every 100<sup>th</sup> was saved into a file. Plots of likelihood were examined for each run to determine the number of generations required to reach stationarity (burn-in) by Tracer v.1.0. (<http://tree.bio.ed.ac.uk/software/tracer/>). Then, the MCMC convergence was evaluated by performing cumulative and sliding window analyses of posterior probability and among-run variability of cumulate and split frequencies using the online application AWTY (Nylander et al. 2008). The initial 2000 trees were discarded. Using the “sumt” command of MrBayes, the 50% majority-rule consensus tree was calculated from 36,000 trees sampled after reaching likelihood convergence to calculate the posterior probabilities of the tree nodes. The statistical congruence among the different regions was tested using ILD test (Farris et al. 1994; Huelsenbeck et al. 1996) carried out with PAUP. A conflict between ITS and *rpb2* and ITS and *ef1α* was found. The incongruities detected among the different data sets appeared in the *C. rei* clade. When incongruities appear among the different data sets, these sets can be analyzed as a whole or separately. This work followed the methodology proposed by Wiens (1998), who advises to separately analyze each data set and to assess the support of each clade; then to carry out a combined analysis of all the data sets, finally deeming as questionable those parts of the tree where incongruities are found.

## Results

### Phylogenetic analyses

In this work, 80 new sequences have been generated, of which 32 are of ITS rDNA, 22 of *rpb2*, and 26 of *ef1α*. The alignment of the ITS rDNA region contained 582 positions while the *rpb2* and *ef1α* alignments contained 891 and 612, respectively.

The MP analyses based on ITS rDNA region generated 500 equally parsimonious trees of 127 steps. The likelihood parameters of Bayesian analyses are shown in TABLE 2. Both analyses generated topologically similar trees. The majority Bayesian consensus tree (FIG. 1A) shows three strongly supported monophyletic clades. One clade groups all the specimens delimited as *C. subulata*; another clade includes all the samples identified as *C. rei*; and the third clade comprises the samples of *C. glauca* and *C. cenotea*. Within the *C. rei* clade, two strongly supported subclades appear. In both subclades, the specimens come from different geographical origins (TABLE 1).

The MP and Bayesian analyses based on *rpb2* partial gene display a similar topology (FIG. 1B). The MP analysis generated 500 equally parsimonious trees, 162 steps long. The rest of the parameters, together with the likelihood values of the Bayesian analysis are shown in TABLE 2. As in the ITS rDNA analyses, three strongly supported clades appear, one corresponding to *C. subulata*, another to *C. rei* and a third including *C. glauca* and *C. cenotea*. Only one strongly

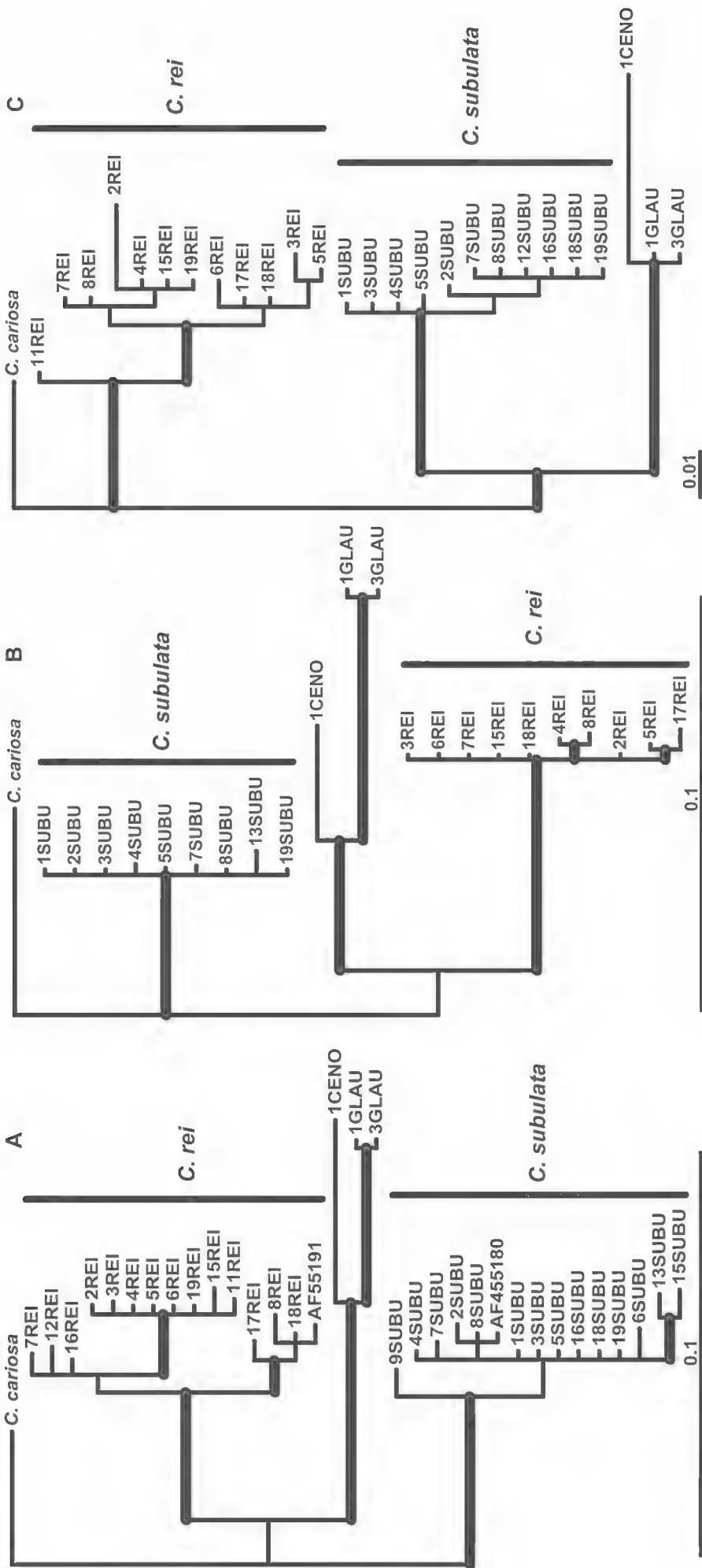


FIG. 1. Phylogeny of *Cladonia subulata* and *Cladonia rei*. The 50% consensus majority-rule tree from Bayesian/MCMC of three separate regions. Bold branches indicate a support of bootstrap  $\geq$  al 70% and posterior probability  $\geq$  95%. A) ITS rDNA B) *rpb2* C) *ef1α*.

supported subclade can be distinguished within the *C. rei* clade. However, it does not correspond to any of those appearing in the ITS rDNA analyses. The samples of this subclade have different geographical origins.

The MP analyses based on *eflα* partial gene generated three equally parsimonious trees of 113 steps. The remaining MP parameters and Bayesian likelihood values are shown in TABLE 2. Analyses corresponding to this *eflα* region also show three strongly supported monophyletic clades (FIG. 1C). In the *C. subulata* clade, one low-support subclade can be observed. The topologies of the MP and Bayesian consensus trees were not strictly identical. The MP tree shows *C. cenotea* apart from the *C. glauca* samples, while the Bayesian tree does not. The Bayesian analysis was repeated using GTR+I+G model and the result was the same.

TABLE 2. Information on MP analyses, evolutionary model and likelihood parameters of Bayesian analyses.

	PARAMETER	ITS rDNA	rpb2	eflα	Combined
MP	CI	0.8920	0.8377	0.9292	0.8667
	RI	0.9530	0.9448	0.9815	0.9518
	RC	0.8501	0.7915	0.9120	0.8249
	informative characters	73	98	66	232
Bayesian analyses	Model	SYM+I	SYM+G	SYM+G	GTR+I+G
	-LnL	-1582.984 (0.07398)	-2128.058 (0.02592)	-1527.70 (0.00723)	-5172.63 (0.01232)
	π (A)	-	-	-	0.2601 (0.00008)
	π (C)	-	-	-	0.2519 (0.00008)
	π (G)	-	-	-	0.2415 (0.00008)
	π (T)	-	-	-	0.2465 (0.00007)
	r (A-C)	0.4745 (0.00235)	0.0468 (0.00031)	0.0605 (0.00051)	0.0591 (0.00016)
	r (A-G)	0.2498 (0.00248)	0.2375 (0.00165)	0.2485 (0.00246)	0.2472 (0.00067)
	r (A-T)	0.1474 (0.00130)	0.0873 (0.00055)	0.0808 (0.00073)	0.1150 (0.00028)
	r (C-G)	0.0666 (0.00050)	0.0326 (0.00022)	0.0312 (0.00036)	0.3326 (0.00009)
	r (C-T)	0.4095 (0.00311)	0.5272 (0.00246)	0.5026 (0.00349)	0.4944 (0.00093)
	r (G-T)	0.0566 (0.00046)	0.0684 (0.00044)	0.0760 (0.00080)	0.0608 (0.00017)
	α	-	0.2748 (0.01024)	0.3535 (0.04190)	73.254 (0.00007)
	Pinvar	0.6021 (0.00198)	-	-	0.6233 (0.00795)

Bayesian parameters: mean value (variance)  
Models selected by AIC criterion using MrModeltest

The MP analyses based on the combined dataset generated 500 equally parsimonious trees of 405 steps long. The remaining parameters of the MP analyses, together with the likelihood values of the Bayesian analyses are shown in TABLE 2. Both analyses generated topologically similar trees (FIG. 2). Three strongly supported monophyletic clades appear, one corresponding to *C. subulata*, another to *C. rei*, and the third to *C. glauca* and *C. cenotea*.

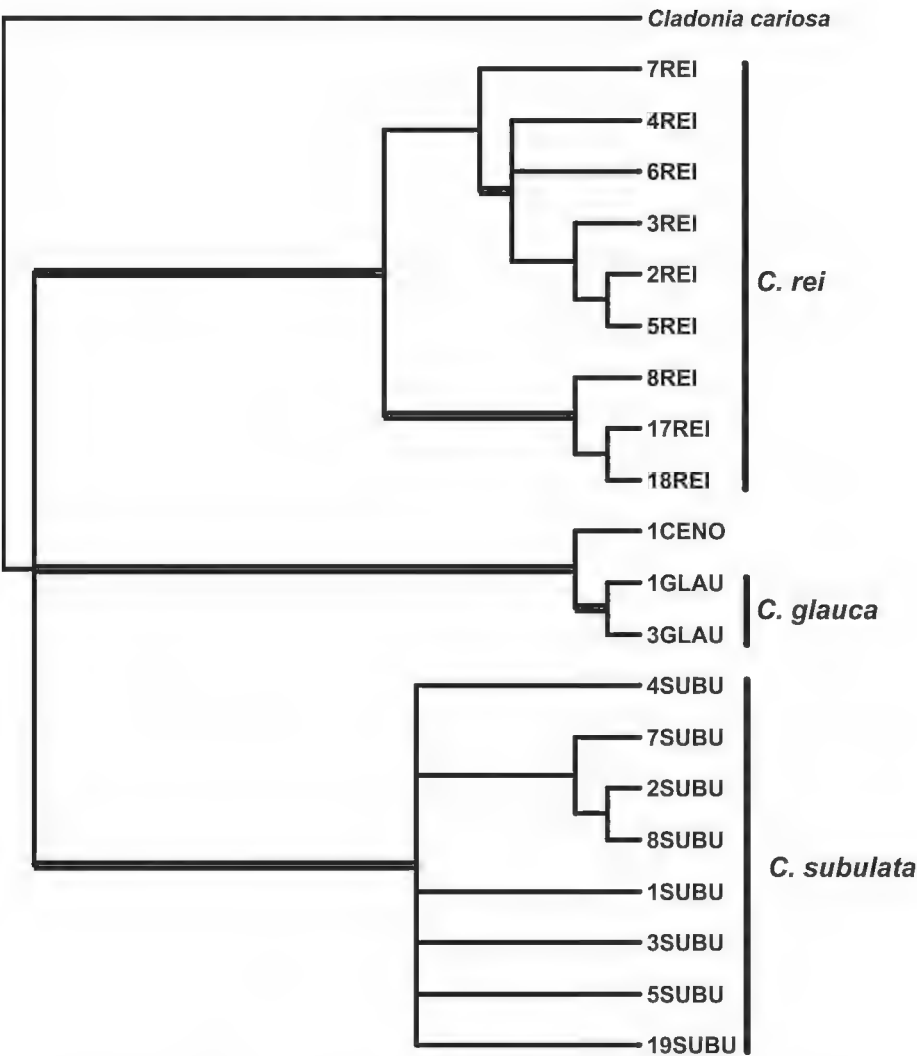


FIG. 2. The 50% consensus majority-rule tree based of combined data set (ITS rDNA, *rpb2* partial gene and *eflα* partial gene) from Bayesian/MCMC. The highly supported branches (bootstrap  $\geq$  70% and posterior probability  $\geq$  95%) are indicated in bold.

The ILD-based congruence analysis revealed one conflict between the ITS rDNA + *rpb2* partial gene matrices and another conflict between the ITS rDNA + *eflα* partial gene matrices. The cause of these incongruities lies in 4 samples of *Cladonia rei* (4REI, 8REI, 17REI and 18REI), which appear in different subclades in the analyses. The three data matrices were combined, however, in accordance with Wiens (1998).

**Morphological and chemical analysis**

The SEM showed notable differences between the stereome surfaces of *Cladonia subulata* and *C. rei*. In *C. rei*, the internal face of the stereome lacks pores, while *C. subulata* samples display a reticulated stereome with pores (FIG. 3). Furthermore, under the light microscope the transverse and lengthwise podetial sections (FIG. 4) reveal stereome hyphae that are thinner in *C. subulata* (2-3  $\mu$ m diam.) than in *C. rei* (3.75-5  $\mu$ m diam.). In both cases, the stereome hyphae are arranged lengthwise along the podetia.



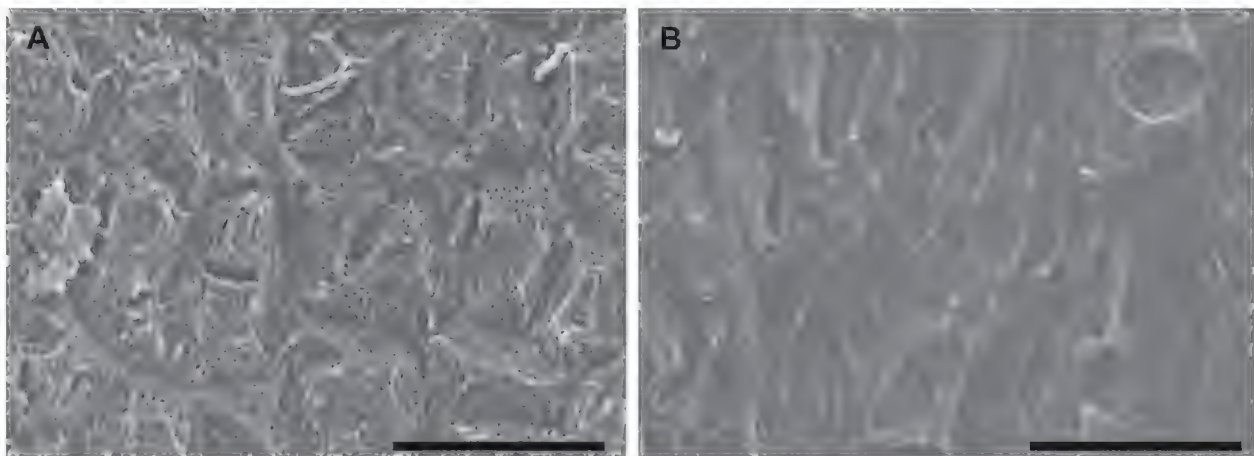


FIG. 3. SEM micrographs of the stereome surface.  
A) *Cladonia subulata*. B) *C. rei*. Bar = 100  $\mu$ m.

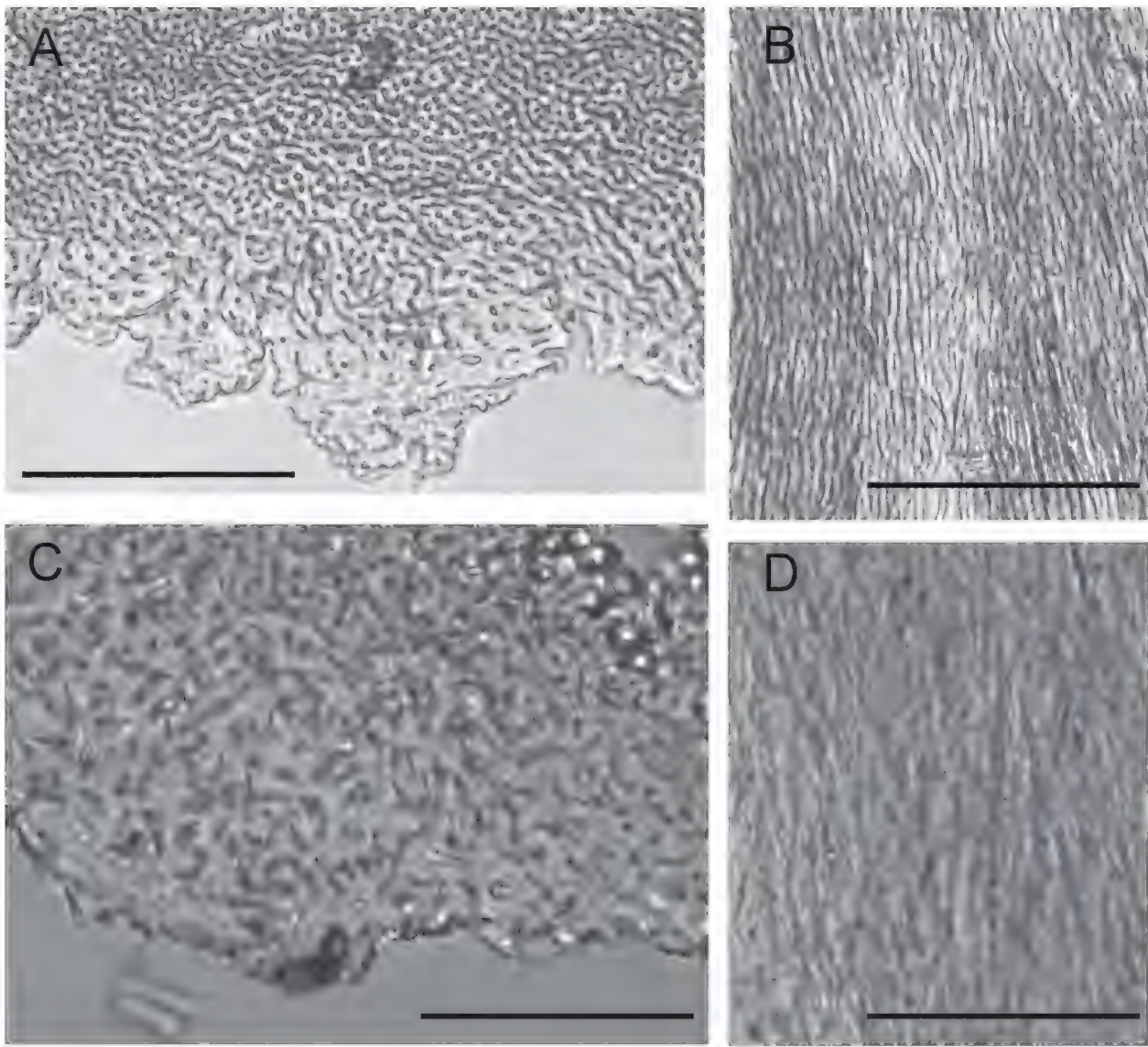


FIG. 4. Microtome sections of stereome under light microscope.  
A) Transversal section of *C. subulata*. B) Lengthwise section of *C. subulata*.  
C) Transversal section of *C. rei*. D) Lengthwise section of *C. rei*.  
Bar = 50  $\mu$ m.

TABLE 3. Results of the contingency table for *C. subulata* and *C. rei*.

CHARACTER	<i>p</i>
Presence/absence of basal squamules	0.035 *
Presence/absence of scyphi	0.13
Presence/absence of basal cortex	0.00008 **
Branching type I/branching type II	0.196

*p*, significance level (\* *p* < 0.05, \*\* *p* < 0.01).

The contingency table (TABLE 3) shows the correlation between the qualitative morphological characters previously used to distinguish these taxa and the clades implied by the phylogenetic analyses. Significant differences are observed, such as the presence/absence of squamules and the presence of basal cortex on the podetia, while there are no significant differences between both taxa in the podetial branching type. Significant statistical differences were found in the podetial anatomical characters (TABLE 4), with the podetial wall being thicker in *C. rei* than in *C. subulata*, as also the medulla and stereome layers are, with the stereome/medulla ratio higher in *C. subulata*. Also, the soredial granules are significantly larger in *C. rei* than in *C. subulata*.

TABLE 4. Statistical analyses for continuous characters.

CHARACTER	<i>C. subulata</i>	<i>C. rei</i>	<i>p</i>
Soredium size	17.5-80 (125)	(14.5) 20-65 (100)	4.42e <sup>-8**</sup>
Podetium thickness	115-310 (350)	(112.5) 130-400 (707.5)	0.0054**
Medule thickness	47.5-225 (250)	(22.5) 30-227.5 (260)	0.0029**
Stereome thickness	35-145 (187.5)	(14.5) 20-212.5 (400)	0.0000**
stereome/medule ratio	1.36-5.0 (5.70)	(1.22) 1.27-2.63 (3.08)	6.57e <sup>-11**</sup>

The minimum value corresponds to percentile 1 and the maximum to percentile 95. The absolute maximum and minimum values are in brackets.

*p*, significance level (\* *p* < 0.05, \*\* *p* < 0.01).

TLC analyses revealed that 36 samples of *C. rei* contained homosekikaic acid together with fumarprotocetraric acid, while 24 samples contained only homosekikaic acid. In both cases, homosekikaic acid was accompanied by small amounts of sekikaic acid. Furthermore, in the samples of *C. rei* the accessory substance 4'-O-methylnorhomosekikaic acid was found. Frequently fumarprotocetraric acid is accompanied by protocetraric acid; besides, in 8 of the samples containing fumarprotocetraric acid, also confumarprotocetraric acid was detected. In all *C. subulata* samples fumarprotocetraric acid was present with protocetraric acid. In addition, in 34 of these samples the satellite substance confumarprotocetraric acid occurred.

The UV test, traditionally used to detect the presence of homosekikaic acid, was applied to 60 samples; 87.5% of the samples where TLC detected homosekikaic acid gave a positive fluorescence. On the other hand, 96%

of the samples where TLC detected only fumarprotocetraric acid gave no fluorescence. The  $\text{FeCl}_3$  test applied to 188 samples gave a positive reaction in 90% of the samples containing homosekikaic acid and was negative in 98% of the specimens containing only fumarprotocetraric acid.

## Discussion

### Evaluation of characters

**SOREDIIUM SIZE.** Soredium size is one of the main characters used for species differentiation in many *Cladonia* species, as in the complex *C. chlorophaea* (Flörke ex Sommerf.) Spreng.–*C. fimbriata* (L.) Fr. (Hennings 1983). However, in *C. ochrochlora* Flörke the soredium size is variable (Hammer 1993). Statistically significant differences in soredium size were found in *C. subulata* and *C. rei*, with the soredial granules being bigger in *C. rei* (TABLE 4). As several factors (e.g., age, development stage, environmental conditions) probably affect soredium size (Paus et al. 1993), using this character to distinguish these species must be used with caution.

**CORTEX AT THE BASE OF PODETIA.** Earlier authors have discussed the utility of the podetial cortex to differentiate *C. rei* from *C. subulata*. Paus et al. (1993) and Spier & Aptroot (2007) consider it unreliable, while Syrek & Kukwa (2008) accept it as reliably diagnostic. Although a great many of the *C. rei* specimens studied were corticated, 40.62% of the *C. subulata* podetia also have corticate bases. The presence of this cortex was sometimes difficult to observe because it was covered by soredia and could be detected only by a transversal section of the podetium.

**SQUAMULES AT THE BASE OF PODETIA.** There are statistically significant differences between the *C. subulata* and *C. rei* clades related to the presence of squamules at the base of podetia (TABLE 4). However, as only 34.69% of *C. rei* podetia have squamules, possession of squamules cannot be used to differentiate these two species. In fact, Evans (1930) differentiated two forms of *C. nemoxyna* (Ach.) Arnold (a synonym of *C. rei*): *C. nemoxyna* f. *fibula* (Ach.) Vainio—lacking podetial squamules—and *C. nemoxyna* f. *phyllocephala* Arn.—with squamulose podetia. The presence/absence of squamules on the podetia is actually a variable character in many *Cladonia* species, e.g., *C. furcata* (Huds.) Schrad. and *C. rangiformis* Hoffm. (Burgaz & Ahti 2009).

**MORPHOLOGY OF PODETIA.** The presence of antler-like, irregularly branched podetia is one character attributed to *C. subulata* (Brodo 2001, Osyczka 2006, James 2009). In the material used for this paper, however, no significant differences were found between the podetia of *C. subulata* and *C. rei*. It is worth noting that much *C. subulata* material studied here was young and not well developed. Other authors (Paus et al 1993, Spier & Aptroot 2007) consider the



podetia morphology to be of little taxonomic value due to the wide variability (simple, cup-like, irregularly branched) that podetia show.

**ANATOMICAL CHARACTERS.** Statistically significant differences between *Cladonia subulata* and *C. rei* were found in the thickness of the podetial wall (TABLE 4). Nevertheless, as in soredium size, the thickness of the podetial wall and the thickness of each layer are widely variable in these two taxa, making it difficult to identify the two species based only on these characters. On the other hand, such anatomical features can be used to differentiate other similar taxa such as *C. mediterranea* P.A. Duvign. & Abbayes from *C. mitis* Sandst., *C. ciliata* Stirt. var. *ciliata* from var. *tenuis* (Flörke) Ahti (Burgaz & Martínez 2008), or the species within the *C. gracilis* (L.) Willd. group (Ahti 1980). In some cases, some taxonomic value is attributed to the stereome surface (Ahti 1980), which is different in *C. rei* and in *C. subulata*. Under the stereomicroscope, the reticulated stereome surface of *C. subulata* and the smooth stereome surface of *C. rei* can sometimes be observed. In most cases, however, a SEM is required to observe stereome surfaces, greatly limiting its utility for an everyday identification. Besides, the differing stereome hyphal thicknesses in those species may be responsible for the differences seen on the stereome surface.

**COLOR OF THE PODETIA.** The color of the podetia of *C. subulata* reportedly varies from whitish-greyish to bright green, up to brownish green, or at least with zones of brownish coloring, while in *C. rei* the podetia vary from brownish green to dirty brown (Suominen & Ahti 1966, Thomson 1968, James 2009); nevertheless color could turn out to be an ambiguous character due to the variation within either species (Paus et al. 1993, Spier & Aptroot 2007). In the present study we found that the podetia of *C. subulata* are often pale green or whitish (though some of them present brownish zones), while in *C. rei* they are green brownish.

**CHEMISTRY.** Secondary metabolites were confirmed as the only reliable characters to distinguish *C. rei* and *C. subulata*. A negative *p*-phenylenediamine (Pd) reaction is still useful in diagnosing specimens as *C. rei*. But a positive reaction is not reliable (Pišút 1961, Paus et al. 1993, Spier & Aptroot 2007), because many *C. rei* samples contain fumarprotocetraric acid in addition to homosekikaic acid, although Suominen & Ahti (1966) note that the *C. rei* Pd reaction is slow, being yellow at first, while in *C. subulata* it is normally instantly red, due to different fumarprotocetraric acid concentrations. Specimens containing homosekikaic acid do appear white under UV, but our results have shown small errors occur in detecting the presence of homosekikaic acid using the UV test. Nonetheless, we find the UV test useful in differentiating the species in most cases. Homosekikaic acid can also be detected by the ferric chloride test, which produces a violet spot when it is positive (Huneck &

Yoshimura 1996). Although this reaction is not used in the keys, we consider it useful for differentiating *C. rei* from *C. subulata*, and it should be included in the identification keys.

### Delimitation of the taxa

Despite the high phenotypic similarity of *C. subulata* and *C. rei*, the phylogenetic analyses of the ITS rDNA, *rpb2* and *efl $\alpha$*  regions show two strongly supported monophyletic clades. These clades agree with the chemical variability of the *C. subulata*-*C. rei* complex. All the specimens included in the *C. rei* clade contain homosekikaic acid with fumarprotocetraric acid as a frequent accessory substance, while in the *C. subulata* clade no specimens with homosekikaic acid were found. If the taxa belonged to a single species with two (to three) chemotypes, it should be expected that the chemotypes would appear intermingled, which is not true. Besides, each clade is associated with a different set of morphological characters.

In addition, the two species have obviously different ecological requirements. *Cladonia rei* is a terricolous species growing in open areas with low humus content and subneutrophilous substrate. It may sometimes grow on impoverished soils with high heavy metal content (Hajdúk & Lisická 1999). *Cladonia subulata* grows on humus-rich acidophilous substrates and even in shady areas (Sipman 1977, Paus et al. 1993, Hammer 1995, Syrek & Kukwa 2008). However, both species do occasionally grow on wood or bare rocks (Spier & Aptroot 2007). Both taxa are broadly distributed in Europe, Asia, and North America and have also been found in Australasia. However, *C. rei* has not been reported for South America or the Antarctic, while *C. subulata* grows in Argentina and Chile. In general *C. subulata* has a wider distribution, although absent in warm areas, while *C. rei* is more common in temperate or sub-arid areas, being absent in Arctic and Antarctic zones (Ahti in litt.).

Suominen & Ahti (1966) found that the *C. rei* chemotypes usually did not appear intermingled, suggesting that the chemotypes are genetically, not environmentally, determined. But the incongruities detected among the different data sets within the *C. rei* clade shows that phylogenetic relationships within this clade are not fully resolved (Wiens 1998).

Our results support *C. subulata* and *C. rei* as two independent phylogenetic species. This conclusion is founded on: 1) the genealogic concordance of the three gene regions; 2) the existence of a correlation between clades and morphological characters; and 3) the fact that both species have different habitats. Our data corroborate the results obtained in the phylogenetic study of *Cladonia* by Stenroos et al. (2002) and Dolnik et al. (2010) where *C. subulata* and *C. rei* appear in separate clades. Spier & Aptroot (2007) pointed out that the Canadian specimen of *C. rei* (AF455191) analyzed by Stenroos et al. (2002)



possibly belongs to another taxon than the European ones. Our ITS analysis, which included this sequence, shows it grouping with the other *C. rei* samples.

*Cladonia glauca* is morphologically similar to *C. rei*, sharing grey brownish podetia and squamules at the podetia base (Brodo et al. 2001, Syrek & Kukwa 2008, James 2009, Burgaz & Ahti 2009). However, they contain different lichen substances representing different biosequential groups. *Cladonia glauca* has squamatic acid or (rarely) thamnolic and barbatic acids (Burgaz et al. 1999, Burgaz & Ahti 2009). In addition, *C. glauca* presents a very peculiar groove along the podetium that distinguishes it from *C. rei*, and it is fully unable to produce cups (scyphi), which occur in mature specimens of *C. rei* and *C. subulata*. Our phylogenetic analyses clearly separate *C. glauca* from *C. rei*. *Cladonia glauca* seems to be related to *C. cenotea* (in some areas they can be difficult to distinguish), and Stenroos et al. (2002) cite *C. cenotea* as phylogenetically related to *C. crispata* (Ach.) Flot. and *C. subsubulata* Nyl. Nevertheless, further studies including additional taxa are necessary to establish the phylogenetic relationships of *C. glauca*.

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## MYCOTAXON

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***Athelopsis parvispora* (Basidiomycetes),  
a new species from India**

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**Abstract** – A new corticioid species, *Athelopsis parvispora*, is described from Manali hills in Himachal Pradesh.

**Key words** – Kullu, Gulaba, stalked basidia

While conducting the fungal forays in the oak forest in Gulaba area of Manali hills district Kullu of Himachal Pradesh, India, Avneet and Dhingra collected a corticioid specimen on a stump of *Quercus incana*. After detailed macroscopic and microscopic comparisons with descriptions of known species of genus *Athelopsis* (Jülich 1971, Eriksson & Ryvarden 1973, Hjortstam 1991, Kotiranta & Saarenoksa 2005), it was found to be close to *Athelopsis subinconspicua* (Litsch.) Jülich. Characters in common were thin, pellicular basidiocarps with smooth hymenial surface and clavate, basally stalked basidia, but the basidiospores in the newly described species differed in being narrowly ellipsoid and smaller ( $4.3\text{--}4.7 \times 1.5\text{--}1.9 \mu\text{m}$ ) compared to the more broadly ellipsoid and larger ( $6.5\text{--}8 \times 4\text{--}4.5 \mu\text{m}$ ) spores in *A. subinconspicua*. This suggests that this new finding represents a species of its own.

***Athelopsis parvispora*** Avneet P. Singh, Dhingra & J. Kaur, sp. nov.

FIGS 1–4

MYCOBANK MB517012

*Basidiocarpum resupinatum, adnatum, effusum, ad 160  $\mu\text{m}$  crassum, pelliculosum; hymenium laevigatum flavescens; systema hyphale monomiticum; hyphae ad 3.1  $\mu\text{m}$  latae, nodoso-septatae; hyphae basales crassitunicatae, incrustatae; subhymenial hyphae tenuitunicatae, non incrustatae; basidia  $10.9\text{--}16 \times 3.0\text{--}3.9 \mu\text{m}$ , clavata, stipitata, 4-sterigmata, ad basin fibuligera; basidiosporae  $4.3\text{--}4.7 \times 1.5\text{--}1.9 \mu\text{m}$ , anguste ellipsoideae, tenuitunicatae.*

TYPE: India, Himachal Pradesh: Kullu, Gulaba, on the way to Rohtang, on *Quercus incana* wood, Avneet 3578 (PUN, holotype), September 10, 2004.

ETYMOLOGY: The epithet refers to small basidiospores.

Basidiocarps resupinate, adnate, effused, up to 160  $\mu\text{m}$  thick in section, thin, pellicular, almost athelioid; hymenial surface smooth, pale yellowish; margins indeterminately thinning. Hyphal system monomitic; generative hyphae up to 3.1  $\mu\text{m}$  wide, branched, septate, clamped; basal hyphae somewhat thick-walled, encrusted; subhymenial hyphae thin-walled, without encrustation. Basidia 10.9–16  $\times$  3.0–3.9  $\mu\text{m}$ , clavate, basally stalked, 4-sterigmate, with a basal clamp; sterigmata up to 4.3  $\mu\text{m}$  long. Basidiospores 4.3–4.7  $\times$  1.5–1.9  $\mu\text{m}$ , narrowly ellipsoid, thin-walled, smooth, inamyloid, acyanophilous.



FIGS 1–4. *Athelopsis parvispora*.

FIGS 1–3. Microscopic structures: 1. Basidiospores; 2. Basidia; 3. Generative hyphae.

FIG. 4. Basidiocarp showing hymenial surface.

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## MYCOTAXON

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***Helicogonium fusisporum* sp. nov.,  
an intrahymenial parasite in *Orbilia eucalypti***

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**Abstract** — *Helicogonium fusisporum*, an intrahymenial ascomycete that forms its ascogenous hyphae and asci in the hymenium of *Orbilia eucalypti* (= *O. coccinella* s. auct. = *O. alnea*), is illustrated and described as a new species from Lithuania.

**Key words** — ascomycetes, taxonomy, mycoparasite

## Introduction

Species of the genus *Helicogonium* W.L. White live as parasites in hymenia of other fungi where they suppress the formation of the host's meiosporangia. They are considered to originate phylogenetically from *Helotiales* but with a loss of ability to form apothecia (Baral 1999).

These species can hardly be detected other than by accident. Although they are certainly not very rare, it is very difficult to search intentionally for them because their presence in the fruitbodies of their hosts (ascomycetes and basidiomycetes) is generally not obvious by external view, and their occurrence is usually irregular and unpredictable. Probably, *Helicogonium* asci have been repeatedly observed by mycologists who put the material aside because the strange combination of ascus and apothecial characters did not fit any described species.

Most species of *Helicogonium* occur as parasites of various genera in the *Helotiales*, whereas only one species, *H. orbiliarum* Baral & G. Marson, was

formerly known to occur in members of *Orbiliomycetes* (Baral 1999). The new species described here is the second one to be found in hymenia of the genus *Orbilia* Fr. (but a third one is mentioned below). It has so far been detected only once, and in spite of a thorough search in Lithuania over a two-year period in more than 30 collections of *Orbilia*, the second author did not succeed in finding it again. Also, the examination of roughly 4500 specimens of *Orbiliomycetes* by the first author and about 100 specimens by the second author during a period of over 20 years never brought this parasitic species to light.

### Material and methods

The type material was studied by both authors in the dead state (the sign † refers to this). Freehand sections made with razor blade and also squashed material were mounted in tap water, 5% aqueous KOH, Lugol's solution (IKI) and aqueous Congo Red (CR) for microscopic examination. Line drawings of microscopical structures were made free-hand directly from the microscope. Photos were obtained with a Nikon Coolpix 4500 digital camera held free-hand on the 10x ocular of a Zeiss Standard 20 microscope. The material is deposited in the Herbaria of the Botanische Staatssammlung München (M) and Institute of Botany, Nature Research Centre, Vilnius (BILAS).

### Taxonomic description

*Helicogonium fusisporum* Baral & Kutorga, sp. nov.

FIGS 1–2

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*Ascomata nulla. Asci in hymenio hospitis formati, 30–47 × (6–)6.5–7.5(–8) µm in statu emortuo, cylindraceo-clavati, tunica apicali incrassata, inamyloidea, octospori, breviter stipitati, plerumque bifurcati, non uncinati. Ascosporae fasciculatae, oblique biseriat, fusioideae, rectae, (7.5–)9–11(–12) × 2–2.5 µm in statu emortuo, non-septatae, guttulis magnis impletae, ascoconidiis carentes. Habitatio: in apotheciis Orbiliae eucalypti.*

TYPE: 55°04'71.7" N, 24°23'74.0" E, Upninkai Forest, Jonava district, Lithuania alt. 122 m., on a xeric still-attached *Quercus robur* branch in apothecia of *Orbilia eucalypti* growing on old ascomata of *Colpoma quercinum*, 2.IX.2004, E. Kutorga (HOLOTYPE – M (ex H.B.8533); ISOTYPE – BILAS 42681).

ETYMOLOGY: referring to the fusoid ascospores.

DESCRIPTION — ASCOGENOUS HYPHAE penetrating the medullary excipulum and subhymenium of the host, simple-septate. ASCI (†) 30–47 × (6–)6.5–7.5 (–8) µm, cylindric-clavate to clavate, 8-spored; apex slightly to medium conical, with an apical dome 2–3 µm (immature) or 1–2 µm (mature) thick in KOH, inner surface plane or usually distinctly convex, without apical chamber, IKI–, usually not exceeding the paraphyses of host in height (dead state); stalk short to medium long, medium thick, bifurcate (Y to L-shaped), without croziers. ASCOSPORES (†) (7.5–)9–11(–12) × 2–2.5 µm, fusoid to fusiform, with gradually

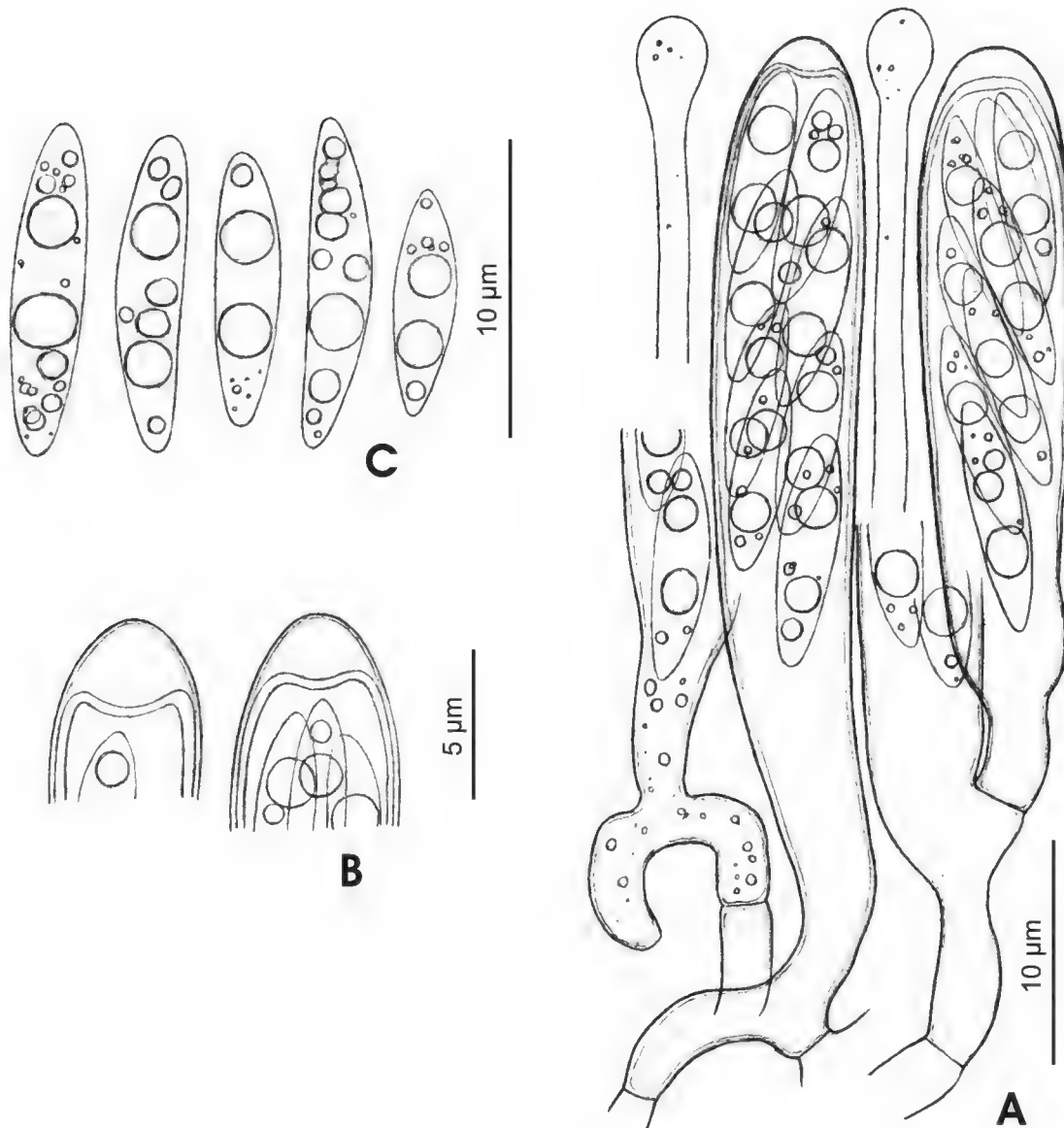


FIG. 1. *Helicogonium fusisporum* (holotype, all in KOH, KOH+IKI or KOH+CR).  
A: asci (between paraphyses of *Orbilia eucalypti*),  
B: ascus apices, C: ascospores containing lipid bodies (oil drops).

tapered, obtuse to acute ends, homopolar, straight to often slightly inequilateral, non-septate, containing some large and small oil drops (high lipid content).

ANAMORPH not detected. Conidia born on ascospores not observed, either on free spores or on spores within the asci.

ECOLOGY, AND RANGE — mycoparasite in the hymenium of *Orbilia eucalypti* (W. Phillips & Harkn.) Sacc., which grew on decayed ascomata of *Colpoma quercinum* (Pers.) Wallr. on 6–9 mm thick, dead, corticated branches attached to a *Quercus robur* tree, ca. 1.5–2 m above the ground in a ca. 70 year old *Pinus sylvestris* stand with scattered *Betula pendula*, *Picea abies*, and *Quercus robur*. So far only known from type locality.



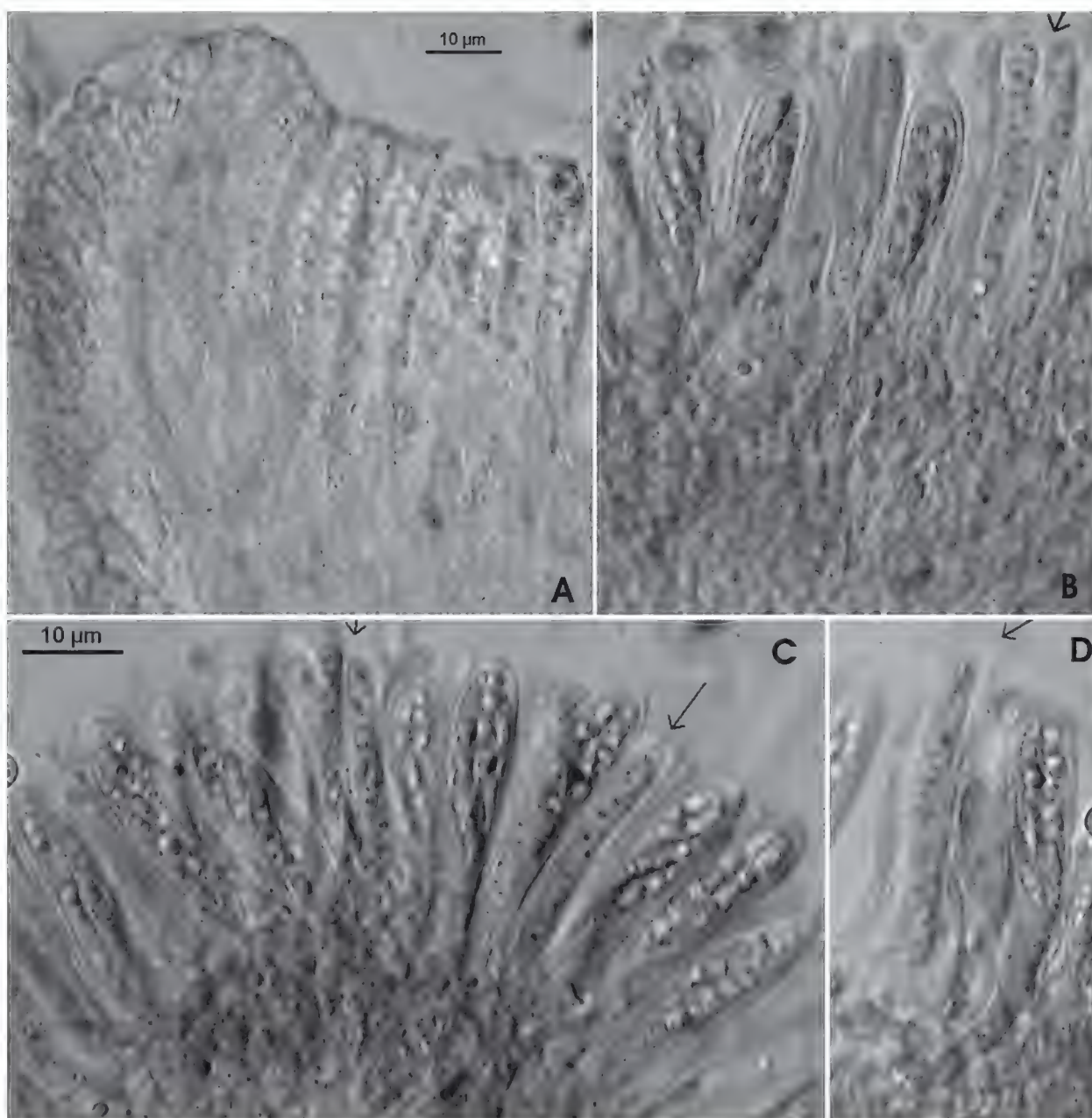


FIG. 2. Asci of *Helicogonium fusisporum* in hymenium of host apothecium (holotype, all in KOH+CR). Arrows: narrow asci of the host *Orbilia eucalypti*.

### Discussion

In 2006, apothecia of *Orbilia eucalypti* (= *Orbilia coccinella* s. auct., = *O. alnea* Velen.) were collected again at the same site on very similar *Quercus* branches with old *Colpoma* ascomata, but no asci of this parasite could be detected in the examined apothecia.

*Helicogonium fusisporum* forms its asci between the paraphyses and asci of the host species *O. eucalypti*. In the apothecia tested, the parasitic asci were present in similar frequency to those of the host. Toward the margin of the apothecia the parasitic asci are fewer. The host asci with their small ellipsoid spores are distinctly narrower than the parasitic asci, while the parasitic asci tend to project more than the host asci (both in the dead state).

*Orbilia eucalypti* is a common species on attached branches or standing trunks in temperate humid to subtropical semi-humid climates. Its apothecia are desiccation-tolerant for at least 1–2 months, but much less tolerant populations of apparently the same species occur on substrate lying on the moist ground. The occurrence of *Helicogonium fusisporum* and its host on rather thin branches at eye height provides evidence that this parasite also is a desiccation-tolerant fungus. Because the specimen was studied only 1–2.5 years after collecting, no observations of the living organs could be made, therefore its desiccation-tolerance is indirectly inferred.

*H. fusisporum* resembles in ascospore shape *H. psilachni* Baral, a parasite in the hymenium of *Psilachnum* aff. *chrysostigmum* (Fr.) Raitv., but in *H. psilachni* the spores are shorter, much more clavate, and produce ascoconidia at their broad end while still inside the immature asci. The type species of *Helicogonium*, *H. jacksonii* W.L. White, parasitic in *Corticaceae*, differs in septate, broader, eguttulate ascospores forming ascoconidia.

Three *Helicogonium* species are presently known to grow parasitically in *Orbilia*. The second, *H. orbiliarum*, is quite common, being so far recorded in seven different species of *Orbilia* (including *O. eucalypti*) as well as in some *Helotiales*, viz. *Calloria* Fr., *Cyathicula* De Not., and *Parorbiliopsis* Spooner & Dennis (Baral 1999). That species is readily recognized by globose to broadly ovoid ascospores containing a few  $\pm$  small lipid bodies, and the spores form small ellipsoid ascoconidia that, prior to ejection, aggregate in 8 “warted” balls within the living mature asci. A third species, *H. cf. hyaloscypharum* Baral, resembles *H. orbiliarum* but differs in more elongate ellipsoid-clavate ascospores producing curved (cashew-shaped) ascoconidia. This species is usually found in hymenia of *Hyaloscypha* Boud. in Europe, although a single known collection from China was detected in an *Orbilia* (*O. cf. crenatomarginata* (Höhn.) Sacc. & Trotter (Hongyan Su pers. comm.)).

During monographic work on the *Orbiliomycetes*, one of us (H.B.) had the opportunity to revise type materials of *O. coccinella* Fr. in Herb. UPS, as well as such of *O. eucalypti* (W. Phillips & Harkn.) Sacc. in K and *O. alnea* Velen. in PRM. It was found that the type of *O. coccinella* possesses 16-spored asci and cashew-shaped ascospores, which is very different from the current concept of that taxon that includes 8-spored asci and ellipsoid ascospores. *Orbilia eucalypti* was found to be conspecific with *O. alnea* and is, therefore, adopted as the oldest available name for *Orbilia coccinella* s. auct., the taxon with ellipsoid ascospores.

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Botanic Gardens at Kew) and Jan Vesterholt (Natural History Museum of Denmark) for reviewing the manuscript.

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***Diadema ahmadii* (Pleosporales),  
a new ascomycetous species from Pakistan**

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**Abstract** — *Diadema ahmadii* sp. nov. is described, illustrated, and compared with similar taxa. This species was collected from dead branches of *Rosa moschata* in Kaghan Valley, an alpine region in Pakistan. *Diadema ahmadii* is most similar to *D. tetramerum*, the type species of the genus, in that it has asci and ascospores of similar dimensions. However, *D. ahmadii* is distinguished from the latter and other related species by having ascospores with a submedian primary septum.

**Key words** — bitunicate ascomycetes, *Diademaceae*, *Dothideomycetes*, *Pleosporomycetidae*

### Introduction

During the examination of several herbarium specimens of bitunicate ascomycetes in Pakistan, an interesting species with dark and relatively large ascospores was found on dead branches of *Rosa moschata* collected from an alpine region in Pakistan (Batakundi, Kaghan Valley). Owing to the presence of globose to subglobose ascomata without a papillate beak, obclavate to cylindrical asci with fissitunicate dehiscence, and deeply pigmented, 3-septate ascospores, this ascomycete was considered as an undescribed species in the genus *Diadema* Shoemaker & C.E. Babc. The new species is described, illustrated, and compared to other species in this genus.

### Materials and methods

Microscopical observations followed methods as described in Tanaka & Harada (2003) and Tanaka et al. (2009) have been followed. To observe the internal structure of ascospores, 5% NaClO was used to bleach strongly melanized ascospores as described in Eriksson (1989). Ratios indicating ascospore septum

**NOTE** — MYCOTAXON prepared this PDF with color plates for the author. The original print version was published with halftone (grayscale) plates.

position follow Shoemaker (1984; length of upper hemispore/total length of ascospore). Holotype and isotype specimens were deposited in the herbaria of LAH (SHI Mycological Herbarium, University of the Punjab) and HHUF (Hirosaki University), respectively.

## Taxonomy

*Diadema ahmadii* Kaz. Tanaka & S.H. Iqbal, sp. nov.

FIGS. 1–15

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*Ascomata* 240–310  $\mu\text{m}$  alta, 290–500  $\mu\text{m}$  diametro, immersa, erumpentia ad apicem, dispersa vel 2–4 congregata, unilocularia, globosa vel subglobosa, glabrata. Orificinum centrale, planum, non papillatum. Paries ascomatis uniformiter 12–30 crassus. Pseudoparaphyses 2–4  $\mu\text{m}$  latae. Asci (135–)140–185(–193)  $\times$  29–42(–44.5)  $\mu\text{m}$ , obclavati vel cylindrici, apice rotundati, stipitati, fissitunicati, octospori. Ascosporae 37–44(–47)  $\times$  13–16  $\mu\text{m}$ , cum septo primum submedio (0.53), 3-septatae, late fusiformis, crassitunicatae, badiae vel fere nigrae, strato mucoso 2–4  $\mu\text{m}$  lato circumdatae.

**HOLOTYPE:** PAKISTAN. NORTHWESTERN FRONTIER PROVINCE, Lalazar (Batakundi), Kaghan Valley, on dead branches of *Rosa moschata*, 3 September 1967, SHI2007-541, LAH (holotype), HHUF 30004 (isotype).

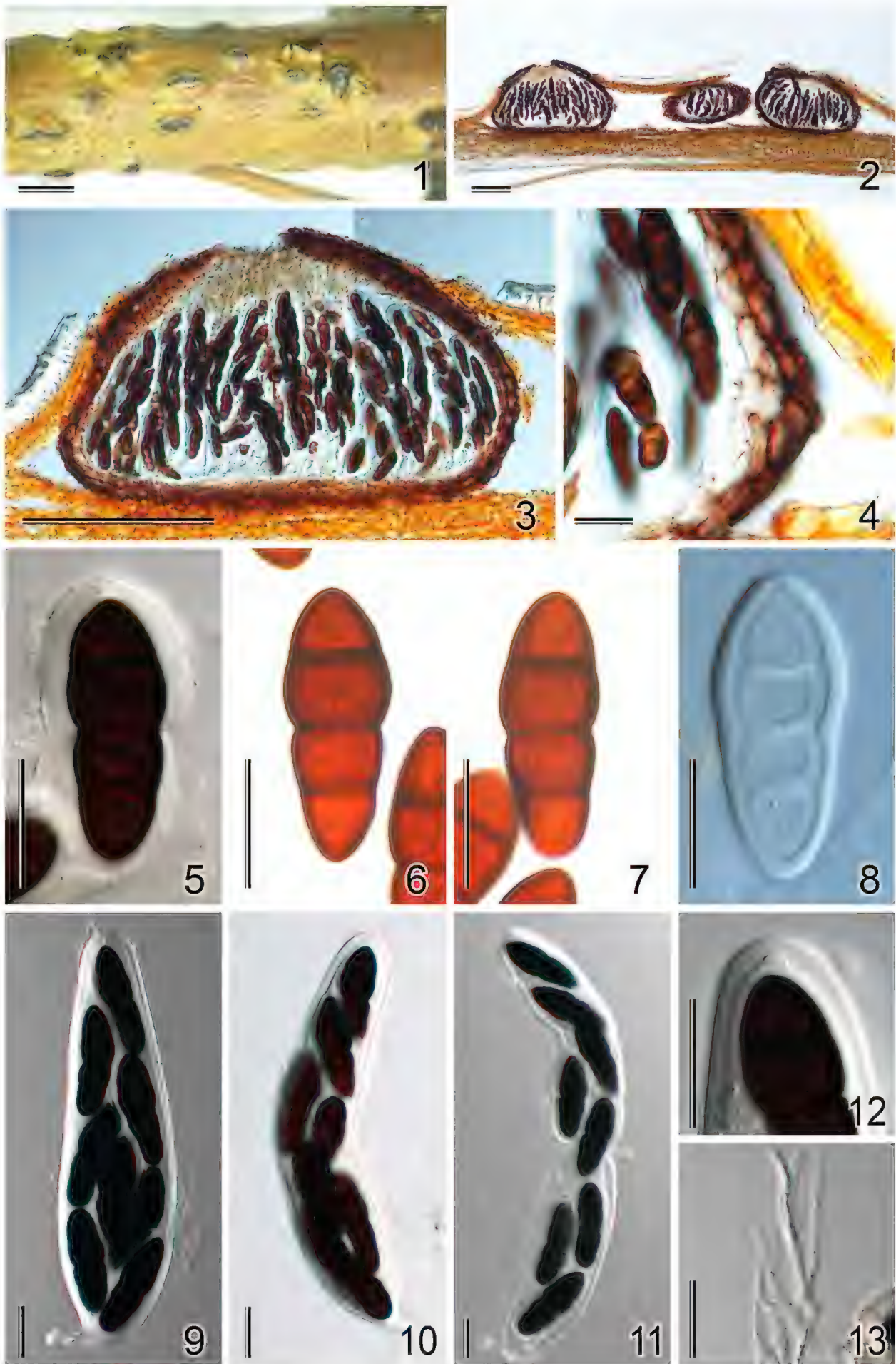
**ETYMOLOGY:** named in honor of Dr. Sultan Ahmad for his outstanding work on Pakistan fungi.

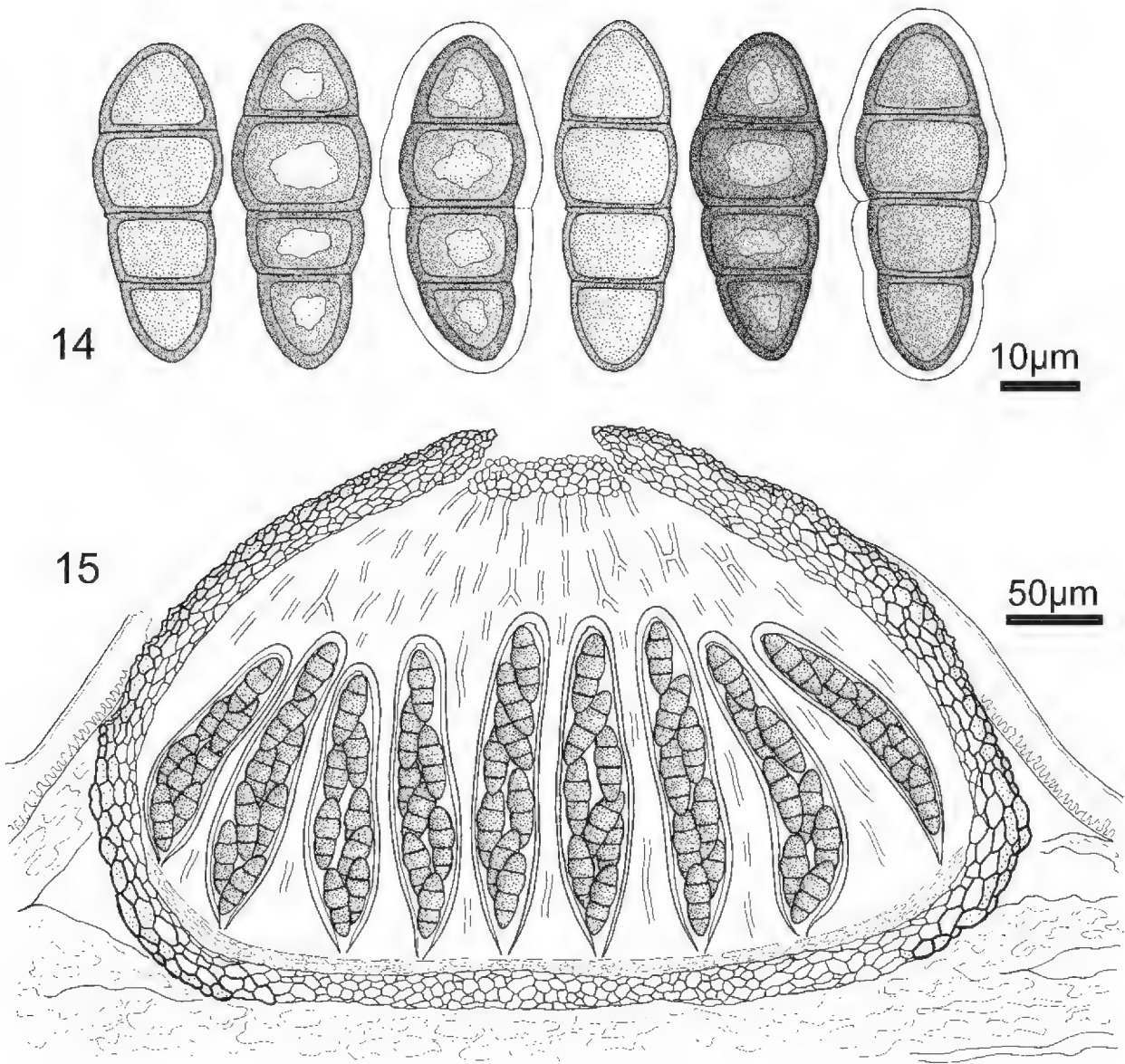
*Ascomata* 240–310  $\mu\text{m}$  high, 290–500  $\mu\text{m}$  diam, immersed, erumpent at the apex, scattered to 2–4 gregarious, uniloculate, globose to subglobose with flattened base in longitudinal section, glabrous. Opening area central, flat, not papillate; lid not seen. Ascomatal wall of ‘textura angularis’ in surface view; wall in longitudinal section uniformly 12–30  $\mu\text{m}$  thick at sides, composed of 4–6 layers of polygonal cells (12–25  $\times$  4–13  $\mu\text{m}$ ); wall at base composed of rectangular to subglobose cells of 5–12  $\mu\text{m}$  diam; wall around the opening area composed of flattened dark brown cells of 5–8  $\times$  2–5  $\mu\text{m}$ . Pseudoparaphyses numerous, sometimes very sparse, septate, branched and anastomosed, 2–4  $\mu\text{m}$  wide. Asci (135–)140–185(–193)  $\times$  29–42(–44.5)  $\mu\text{m}$  (mean = 159.3  $\times$  36.6  $\mu\text{m}$ ,  $n$  = 50), obclavate to cylindrical, rounded at the apex, with a shallow ocular chamber, broadly at below, stipitate, fissitunicate, with 8 biseriate ascospores. Ascospores 37–44(–47)  $\times$  13–16  $\mu\text{m}$  (mean = 41.3  $\times$  15.0  $\mu\text{m}$ ,  $n$  = 70), L/W 2.5–3.1 (mean = 2.8,  $n$  = 70), with a primary septum submedian (0.51–0.55; mean = 0.53,  $n$  = 70), 3-septate, rarely with an additional septum at the basal cell, broadly fusiform, mostly straight, enlarged at second cell from apex, strongly constricted at primary septum, weakly constricted at other septa,

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FIGS. 1–13. *Diadema ahmadii*. 1. Ascomata on the host surface. 2–3. Longitudinal section of ascomata. 4. Ascomatal wall in longitudinal section. 5–7. Ascospores in water mount. 8. Ascospore in NaClO. 9–10. Asci. 11. Extending fissitunicate ascus. 12. Apex of ascus. 13. Pseudoparaphyses. Bars: 1 = 1 mm, 2–3 = 200  $\mu\text{m}$ , 4–13 = 20  $\mu\text{m}$ .







FIGS. 14–15. Line drawings of *Diadema ahmadii*.  
14. Ascospores. 15. Ascoma in longitudinal section. Bars: 14 = 10 μm. 15 = 50 μm.

thick-walled (ca. 1–2 μm wide), reddish brown to almost black, smooth, with a sheath. Sheath entire, firm, sharply delimited, 2–4 μm thick, mostly constricted at the side of primary septum.

### Discussion

*Diadema*, an ascomycetous genus typified by *D. tetramerum* Shoemaker & C.E. Babc., is assigned to *Diademaceae*, a family characterized by the presence of a 'lid' or 'cap' in the area of the ascomatal opening (Shoemaker & Babcock 1992). Most members of *Diadema* are known from culms or stems of alpine plants, in particular, those belonging to *Poaceae* or *Rosaceae*, and are reported from India, Pakistan, and USA (Shoemaker & Babcock 1989). The characteristic features of *Diadema* are relatively large, deeply pigmented ascospores and the disc-like

opening system of the ascomata. These morphological ascomycete features are generally regarded as adaptations to severe alpine conditions, such as high UV-radiation and low temperature (Savile 1972, Leuchtmann 1987, Shoemaker & Babcock 1989). *Diadema* was monographed by Shoemaker & Babcock (1989), who accepted 6 species in this genus. Subsequently, one species was added by Huhndorf (1992).

The overall morphological features of *Diadema ahmadii* as well as its alpine habitat in Pakistan agree with the current concept of *Diadema*. However, we could not find a ‘lid’ or ‘cap’ at the opening of the ascomatal apex in our material. This cap-like structure is also absent in other species of *Diadema*, such as *D. sieversiae* (Peck) Huhndorf and *D. obtusum* Shoemaker & C.E. Babc. (Shoemaker & Babcock 1987, Huhndorf 1992). The opening area of the ascomata in the Pakistan material was obscure, but the presence of a flattened apex in ascomata that lacked a papillate beak, the wall around the opening area composed of small flattened dark brown cells, and the subtending pseudoparaphyses suggest that the opening system of *D. ahmadii* might be essentially the same as that of other species in *Diadema*.

TABLE 1. Comparison of *Diadema ahmadii* with other species in the genus *Diadema*

TAXA	ASCOSPORES				HOSTS	DISTRIBUTION
	Size (µm)	L/W	PS*	Septa		
<i>D. ahmadii</i> (this study)	37–44(–47) × 13–16	2.8	0.53	3	<i>Rosaceae</i>	Pakistan
<i>D. acutum</i> Shoemaker & C.E. Babc. <sup>1)</sup>	50–56 × 16–21	2.9	0.50	3	<i>Poaceae</i>	India, USA
<i>D. cinctum</i> Shoemaker & C.E. Babc. <sup>1)</sup>	33–38 × 13–18	2.3	0.50	3	<i>Cyperaceae</i>	India
<i>D. curtum</i> Shoemaker & C.E. Babc. <sup>1)</sup>	43–49 × 15–20	2.7	0.50	3	<i>Biebersteiniaceae</i>	India
<i>D. hexamerum</i> Shoemaker & C.E. Babc. <sup>1)</sup>	40–50 × 15–17	2.6	0.50	5	<i>Poaceae</i>	USA
<i>D. obtusum</i> <sup>1)</sup>	(40–)46–50(–55) × 19–21	2.3	0.50	3	<i>Rosaceae</i> , <i>Poaceae</i>	India, Pakistan
<i>D. sieversiae</i> <sup>2) 3)</sup>	50–56 × 20–28	–	0.48	3	<i>Rosaceae</i> , <i>Ericaceae</i>	USA
<i>D. tetramerum</i> <sup>1)</sup>	36–50 × 14–20	2.7	0.50	3	<i>Juncaceae</i> , <i>Poaceae</i>	USA

Data from <sup>1)</sup> Shoemaker & Babcock (1989), <sup>2)</sup> Shoemaker & Babcock (1987), <sup>3)</sup> Huhndorf (1992).  
\* PS = position of the primary septum (length of upper hemispore/total length of ascospore).

Among the 7 species previously recognized in this genus, *D. ahmadii* superficially resembles *D. tetramerum*, the type species of the genus, in having asci and ascospores of similar dimensions. Like *D. ahmadii*, the species, *D. obtusum* and *D. sieversiae*, have also been recorded on host plants belonging to the family *Rosaceae* in alpine regions. *Diadema ahmadii*, however, can be easily distinguished from all known *Diadema* species owing to the presence of ascospores with submedian primary septum. A synopsis of these differences is shown in TABLE 1.

### Acknowledgments

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***Ganoderma hoehnelianum* has priority over *G. shangsiense*,  
and *G. williamsianum* over *G. meijiangense***DONG-MEI WANG<sup>1</sup> & SHENG-HUA WU<sup>2</sup>*shwu@mail.nmns.edu.tw*<sup>1</sup>*Guangdong Provincial Key Laboratory of Microbial Culture Collection  
and Application, Guangdong Institute of Microbiology  
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**Abstract** — Some type specimens of *Ganoderma* from tropical and subtropical Asia were studied. The results revealed that *Ganoderma hoehnelianum* and *G. williamsianum* are earlier names for two species of *Ganoderma* originally described from China, *G. shangsiense* and *G. meijiangense*, respectively.

**Key words** — *Elfvingia*, *Ganodermataceae*, *Polyporales*, taxonomy

**Introduction**

In China, Zhao & Zhang (2000) considered the genus *Ganoderma* P. Karst. to contain three subgenera and discriminated subgenus *Elfvingia* (P. Karst.) Imazeki from the other two by its non-laccate upper pilear surface, thick cuticle of trichodermic, anamixodermic, or plecodermic composition and uniformly brown, dark brown, or chestnut brown context. In the same paper they recorded twenty species of subgenus *Elfvingia* from this region and ten of them were new to science (Zhao et al. 1984, 1986; Zhao & Zhang 1986, 1987a,b; Zhao 1988a,b). After studying the type specimens of these “new species,” we found that *Ganoderma meijiangense* and *G. shangsiense* are synonyms of *G. williamsianum* and *G. hoehnelianum* respectively. Descriptions for these two species were based solely on Chinese collections.

Methods for morphological studies mainly followed those previously described by Wang & Wu (2007). Sections for cuticular observations were

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\* Author for correspondence.



taken from the pileus, and color of cuticular hyphae was recorded when treated with Melzer's reagent. Basidiospores were mounted in 5% KOH and only spores with a collapsed apex were measured.

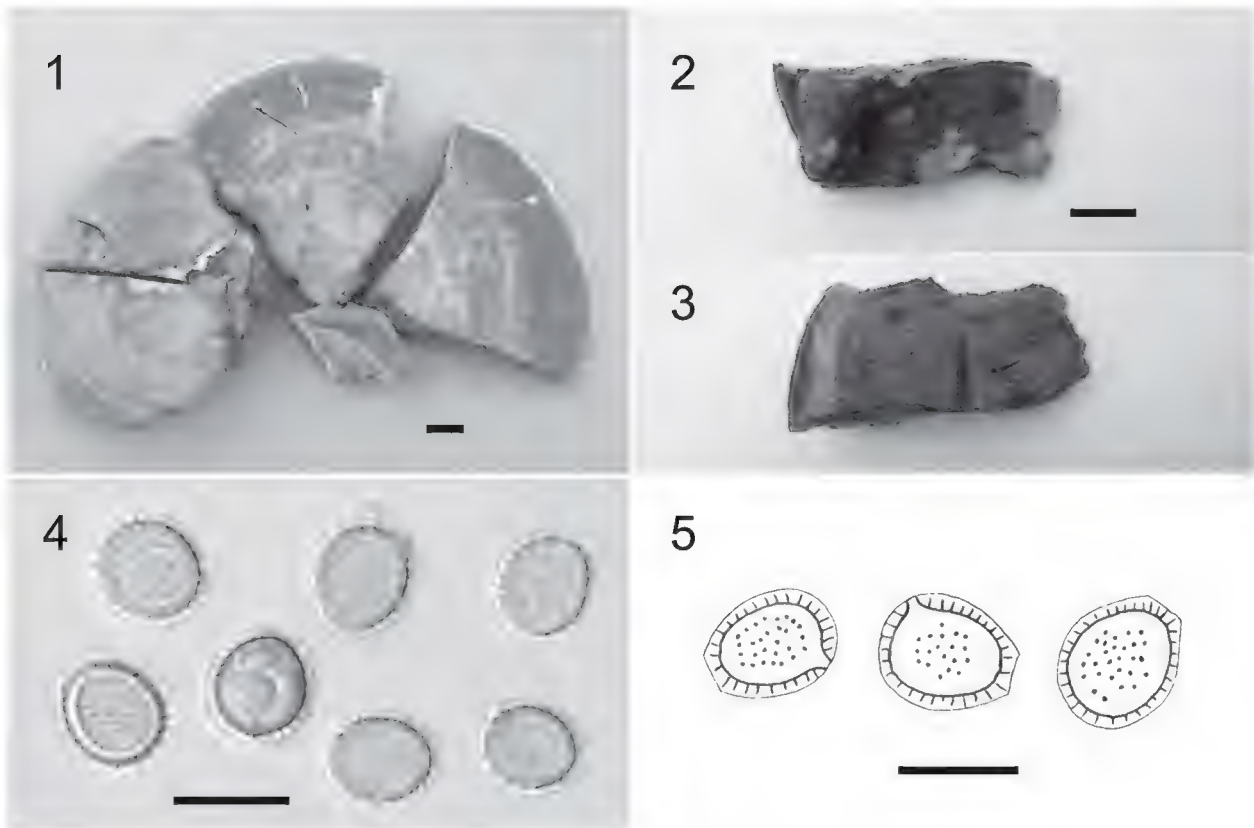
Taxonomy

*Ganoderma hoehnelianum* Bres., Ann. Mycol. 10(5): 502 (1912). FIGS. 1–5  
= *Ganoderma shangsiense* J.D. Zhao, Acta Mycol. Sin. 7(1): 17 (1988).

A full description of Chinese *G. hoehnelianum* was given by Wang et al. (2005; as *G. shangsiense*). The description of *G. shangsiense* was based on its holotype (HMAS 29739) and five other Chinese collections (HMAS 29740, 29741, 29742, 73477, 130043).

SPECIMENS EXAMINED—CHINA. GUANGXI: Shangsi county, on rotten wood, 5 Nov. 1958, Z.-C. Liang 1652 (HMAS 29741). HAINAN: Diaoluoshan, on rotten wood, 29 Sep. 1958, J.-H. Yu 325 (HMAS 29739, **Holotype** of *G. shangsiense*); Diaoluoshan, on dead wood, 25 Sep. 1958, J.-H. Yu 147 (HMAS 29740); Diaoluoshan, on living tree, 4 Oct. 1958, J.-H. Yu 350 (HMAS 29742); Diaoluoshan, on rotten wood of broad-leaved tree, 11 Apr. 1993, J.-P. Lai 1799 (HMAS 73477); Diaoluoshan, on dead wood of *Quercus patelliformis*, 13 Dec. 2003, D.-M. Wang 28 (HMAS 130043). INDONESIA. JAVA: Tjibodas, v. Hoehnel (BPI 236008, **Isotype** of *G. hoehnelianum*).

DISTRIBUTION—Indonesia (Bresadola 1912, Ryvarden 1988), China (this study).



FIGS 1–5. *Ganoderma hoehnelianum* (FIGS 1, 4: HMAS 29739; FIGS 2, 3, 5: BPI 236008). FIG 1. Basidiocarps; FIG 2. Upper surface of the basidiocarp fragment; FIG 3. Pore surface of the basidiocarp fragment; FIG 4. Basidiospores; FIG 5. Basidiospores. Bars = 1 cm in FIGS 1 & 2; = 10 µm in FIGS 4 & 5.

NOTES—Only a small pilear fragment remains from the isotype of *G. hoehnelianum* (BPI 236008). However, this portion was enough to recognize the species. The basidiocarp has a dull yellowish brown to blackish brown upper pilear surface; a vividly yellow pore surface, becoming purplish-brown on bruising; duplex context with yellow or bright yellow approaching the cuticle and yellowish brown to brown near the tube layer, with two black crustose layers; pale brown to brown tubes; broadly ovoid to subglobose basidiospores with thick echinulae and only a slightly truncate apex ( $11.0\text{--}12.0 \times 8.5\text{--}9.5\ \mu\text{m}$ ); an anamixodermic cuticle composed of pale yellow, interwoven hyphae. The combined features of context color, basidiospore characteristics, and cuticular composition are the most reliable criteria in recognizing *G. hoehnelianum*. The Chinese collections of *G. shangsiense* bear the same characteristics (Wang et al. 2005).

*Ganoderma williamsianum* Murrill, Bull. Torrey Bot. Club 34: 478 (1907).

FIGS. 6–12

= *Elfvingia williamsiana* (Murrill) Imazeki, Bull. Gov. Forest Exp. St. Tokyo 57: 106 (1952).

= *Ganoderma meijiangense* J.D. Zhao, Acta Mycol. Sin. 7(1): 16 (1988).

BASIDIOMA annual to perennial, mostly sessile, rarely with a stipe-like base, lightweight, corky. PILEUS  $3.5\text{--}4.7 \times 6.5\text{--}10.0\ \text{cm}$ , reniform, dimidiate, unguulate or irregularly shaped due to imbrication; upper surface reddish brown to purplish black or black, partly strongly laccate, finely but distinctly and concentrically sulcate, slightly to distinctly radially rugose; margin rounded, incurved, concolorous with the pileus. PORE SURFACE dark yellow or bright yellow; tubes up to 1.9 cm long in total, brown or dark brown; pores circular, 5/mm,  $100\text{--}200\ \mu\text{m}$  diam., dissepiments  $45\text{--}90\text{--}(120)\ \mu\text{m}$  thick. CONTEXT up to 1.5 cm thick, yellowish brown to reddish brown, with black crustose layers, corky; generative hyphae  $3.0\text{--}4.5\ \mu\text{m}$  diam., colorless, thin-walled; skeletoligative hyphae  $5.5\text{--}8.0\ \mu\text{m}$  diam., yellowish brown to reddish brown in KOH, arboriform with short sinuous branches. BASIDIOSPORES  $(10.5\text{--})11.5\text{--}13.2\text{--}(14.5) \times (7.5\text{--})8.5\text{--}9.5\text{--}(11.0)\ \mu\text{m}$  (with myxosporium),  $(9.2\text{--})10.0\text{--}11.5\text{--}(12.0) \times (6.2\text{--})7.5\text{--}8.0\text{--}(9.5)\ \mu\text{m}$  (without myxosporium), ellipsoid, mostly truncate at apex, brown, with a dark brown eusporium bearing very thick echinulae and longitudinally ridged ornamentations. CUTIS anamixodermic, composed of yellowish brown, dextrinoid, thick-walled hyphae usually with numerous irregular protuberances, and colorless or pale yellow, thin-walled hyphae arising from the yellowish brown hyphae, easily broken and peeled off.

SPECIMENS EXAMINED—CHINA. HAINAN: Hainan Botanical Garden, alt. 300 m, on rotten wood, 31 Oct. 1958, J.-H. Yu & J.-C. Xing 535 (HMAS 27076); Diaoluoshan, on fallen wood, 6 Nov. 1960, J.-H. Yu & R. Liu 2807 (HMAS 31826, **Holotype** of *G. meijiangense*); Diaoluoshan, on rotten wood, 26 Sep. 1958, R.-Y. Zheng et al. 212 (HMAS 26159); Jianfengling, on dead standing tree, 5 May 1960, J.-H. Yu & R. Liu 1262 (HMAS

30879). YUNNAN: Meijiang county, on rotten wood, 19 Apr. 1957, Bailiangshiji (HMAS 29751). PHILIPPINES. LUZON: Lamao River, Jan. 1904, R.S. Williams (BPI 236684, Isotype of *G. williamsianum*).

DISTRIBUTION—Philippines (Murrill 1907, Steyaert 1972), Indonesia (Imazeki 1952, Steyaert 1972), Malaysia (Steyaert 1972), China (this study).

NOTES—The isotype of *G. williamsianum* (BPI 236684) comprises only a slice of a basidiocarp. Based on this material, the main features of *G. williamsianum* can be summarized as follows: strongly laccate, reddish brown pileus; vividly yellow pore surface; reddish brown context becoming yellowish brown near the cuticle, with two black crustose layers; skeleto-ligative hyphae with consistently short branches; pale brown tubes; ellipsoid basidiospores with rather thick echinulae and longitudinally ridged ornamentations ( $13.5\text{--}16.0 \times 9.0\text{--}10.5 \mu\text{m}$ ); cuticle composed of thin-walled, interwoven hyphae usually with apical protuberances, colorless or pale yellow (inamyloid) or bluish black (amyloid).

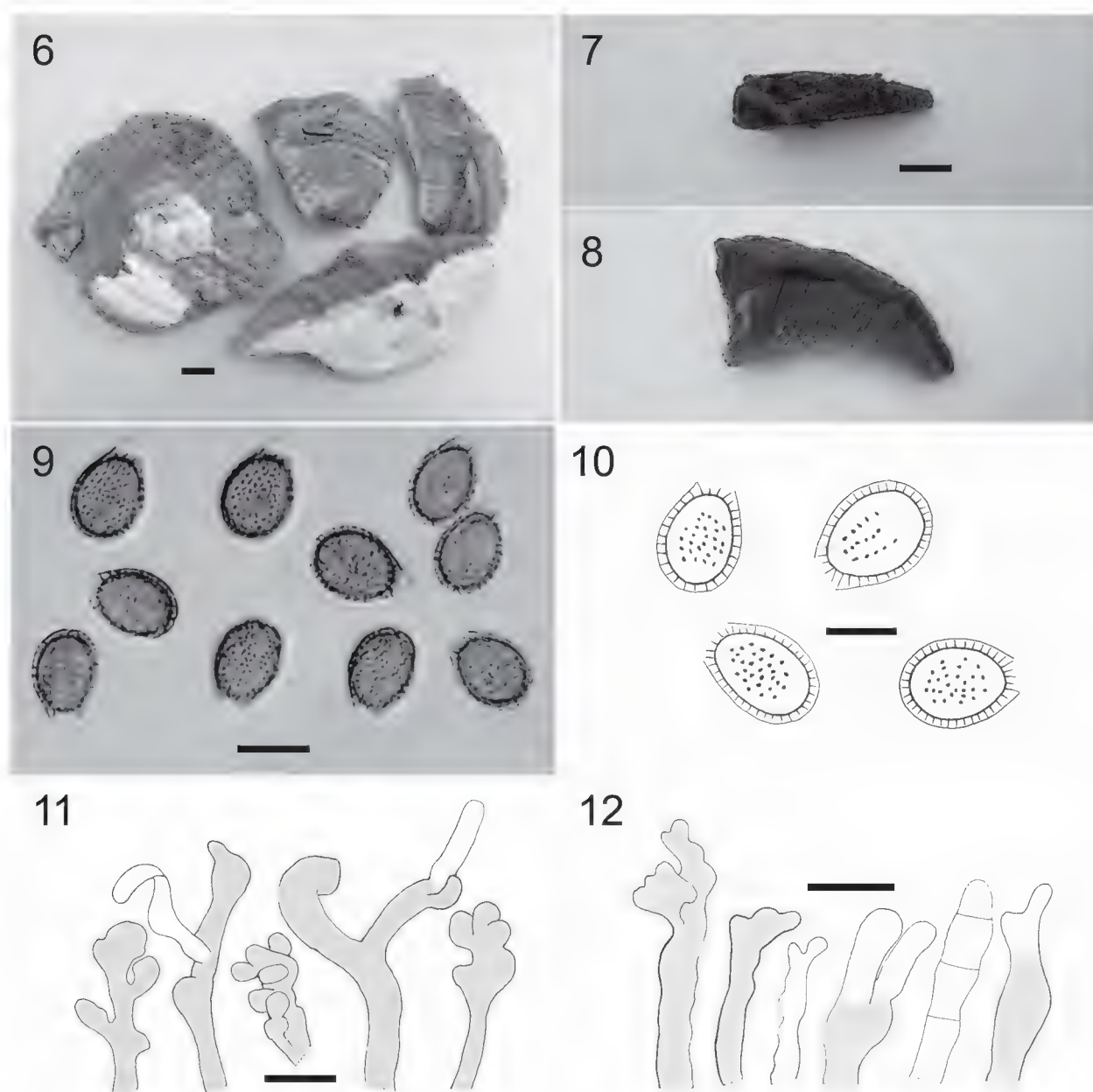
Steyaert (1972) emphasized peculiarities of two characters in *Ganoderma williamsianum*. First, the cutis is composed of “hyaline” hyphae only; secondly, hyphae grow in a wavy or zigzag manner. The first feature is merely one transitional form from anamixoderm to characoderm (Corner 1983). The second feature illustrated in Steyaert (1972) and Corner (1983) is from short sinuous branches at the ends of the skeletal hyphal stalk. In addition, *G. williamsianum* is also easily recognized by having a yellow pore surface, dark brown context, and large basidiospores with striped ornamentation.

*Ganoderma williamsianum* superficially resembles members of the laccate *Ganoderma* group by its macromorphology. Aoshima (1971) misinterpreted the cuticle of this species as a palisadoderm. In reality, *G. williamsianum* has an anamixodermic cuticle and is a member of *Elfvingia* group (Imazeki 1952, Moncalvo & Ryvarden 1997).

Corner (1983) considered that *G. williamsianum* needed to be compared with *G. brownii* (Murrill) Gilb., an American species collected from California. *Ganoderma brownii* is very similar to *G. williamsianum* in color of pore surface (Lowe & Gilbertson 1961, Gilbertson & Ryvarden 1986). However, *G. brownii* can be easily distinguished from *G. williamsianum* by having a dull pileus with a hard, not easily broken crust (Lowe & Gilbertson 1961) formed by hyphae arranged in a trichoderm (Steyaert 1972, Gottlieb & Wright 1999), skeletal hyphae with occasional branching (Gilbertson & Ryvarden 1986), and smaller basidiospores ( $9\text{--}12 \times 7\text{--}9 \mu\text{m}$  in Lowe & Gilbertson (1961),  $9.5\text{--}10.6\text{--}12 \times 6.5\text{--}7.6\text{--}8 \mu\text{m}$  in Steyaert (1972),  $11\text{--}12 \times 7\text{--}8 \mu\text{m}$  in Gilbertson & Ryvarden (1986),  $(9\text{--})10\text{--}11 \times 6\text{--}7(-8) \mu\text{m}$  in Gottlieb & Wright (1999)).

Zhao (1988a) stated that *G. williamsianum* is similar to *G. meijiangense* but distinguished from the latter by having dark brown context without any black crustose layer and a distinct cuticular composition. However, these





FIGS 6–12. *Ganoderma williamsianum* (FIGS 6, 9, 11: HMAS 31826; FIGS 7, 8, 10, 12: BPI 236684). FIG 6. Basidiocarps; FIG 7. Upper surface of the basidiocarp fragment; FIG 8. Vertical section of the basidiocarp fragment; FIG 9. Basidiospores; FIG 10. Basidiospores; FIG 11. Cutis hyphae (Pale parts indicating colorless or pale yellow, thin-walled hyphae; Dark parts indicating yellowish brown, thick-walled hyphae); FIG 12. Cutis hyphae (Pale parts indicating colorless or pale yellow, thin-walled hyphae; Dark parts indicating bluish black hyphae). Bars = 1 cm in FIGS 6 & 7; = 10 µm in FIGS 9–12.

characters used for discrimination by Zhao (1988a) have not been supported in this study. The isotype of *G. williamsianum* BPI 236684 has dark context with black crustose layers, while the holotype of *G. meijiangense* HMAS 31826 has a cuticular composition with colorless or pale yellow, thin-walled hyphal ends. Further, HMAS 31826 has the skeleto-ligative hyphae which are typical for *G. williamsianum*. The Chinese collections of *G. meijiangense* cited above agree well with *G. williamsianum* in morphology except for having slightly smaller basidiospores.

### Acknowledgments

We are very grateful to Drs. Nils Hallenberg and Peter Buchanan for reviewing this paper. Thanks are due to the curators of BPI and HMAS for loans of *Ganoderma* specimens. This study is supported by the National Museum of Natural Science and Foundation of the National Museum of Natural Science of ROC, Postdoctoral Fellowship Grant of National Science Council (NSC96-2816-B-178-001), the Natural Science Foundation of Guangdong Province, China (Serial no. 8451007002001904), Science and Technology Planning Project of Guangdong Province, China (2008B020400013, 2009B020304003), and the Foundation of Guangdong Academy of Sciences, China for 2008 Outstanding Young Science and Technology Talents. The senior author also wishes to appreciate the instructions of Dr. Y.-J. Yao during her initial study of *Ganoderma* in China.

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## MYCOTAXON

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***Passalora acericola* – a rare cercosporoid species  
found for the first time in Poland**

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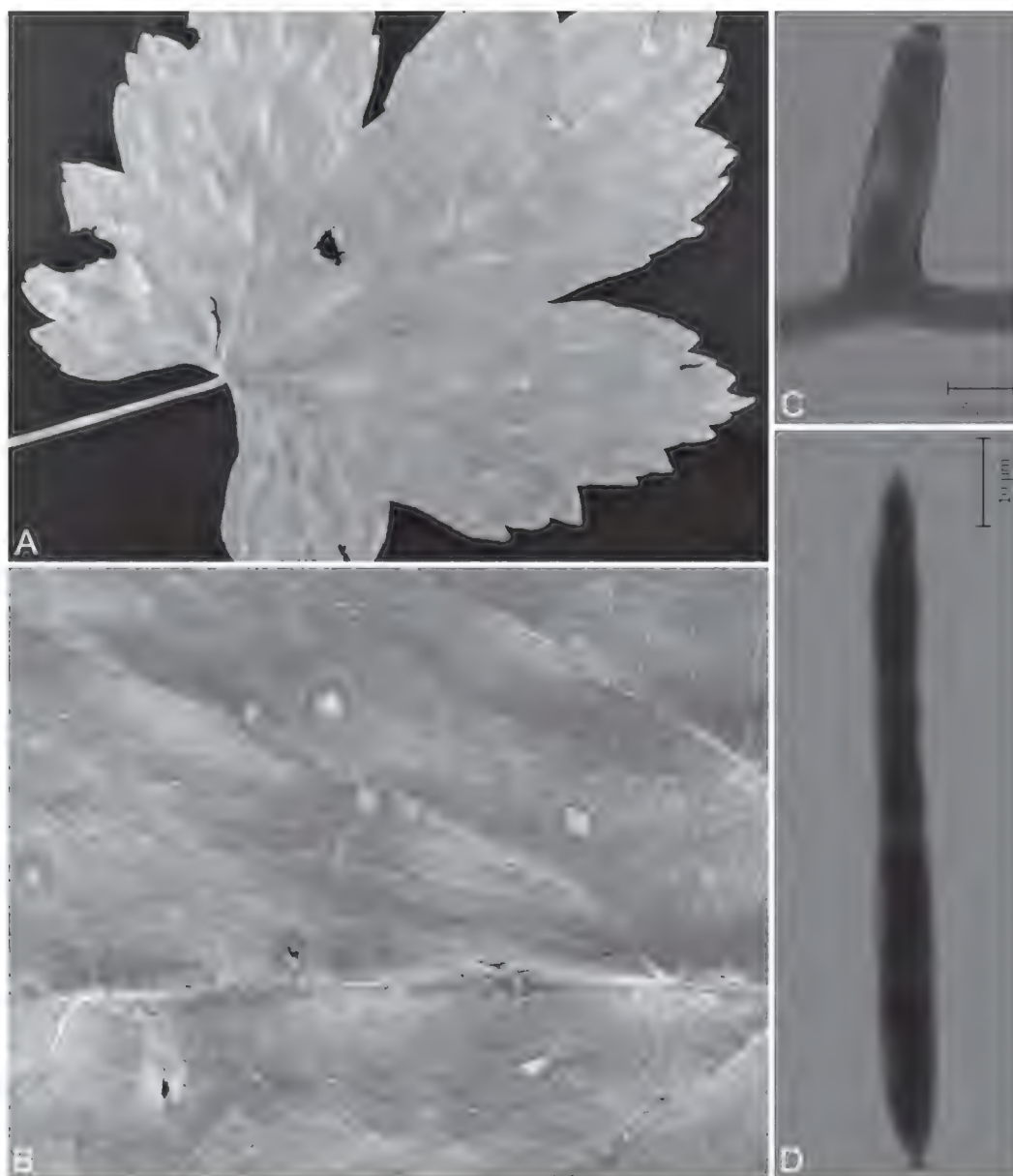
**Abstract** – The rare cercosporoid hyphomycete *Passalora acericola* has been found for the first time in Poland, on *Acer pseudoplatanus*. Previously, this rare species has been found in only three other localities. It is described illustrated and discussed, based on the Polish material.

**Key words** – anamorphic fungi, hyphomycetes, distribution

**Introduction**

*Passalora* Fr. was previously regarded as an anamorph genus of the ascomycetous genus *Mycosphaerella* Johanson (e.g., Braun & Mel'nik 1997, Shin & Kim 2001, Crous & Braun 2003) and belonged to the so-called cercosporoid fungi. *Passalora* is now considered polyphyletic within *Mycosphaerellaceae* and not a genus-specific anamorph of *Mycosphaerella* s. str., which is restricted to species having *Ramularia* anamorphs (Crous et al. 2009). *Passalora*-like fungi are usually phytopathogenic, often causing leaf spots, but they may occasionally also be hyperparasitic or rarely saprobic (Crous & Braun 2003). Fries introduced *Passalora* as a genus in 1849. Braun (1995) discussed in detail the differentiation of *Passalora* and allied genera within the cercosporoid fungi. Recently Crous & Braun (2003) recognized four true cercosporoid genera, viz. *Cercospora* Fresen., *Passalora*, *Pseudocercospora* Speg., and *Stenella* Syd. and cited several other morphologically similar genera based on molecular sequence analyses and a reassessment of morphological characters.

*Passalora acericola* is a very rare species known from only three other localities in the world. Liu & Guo (1982) first described the fungus as *Phaeoramularia acericola* X.J. Liu & Y.L. Guo on *Acer truncatum* Bunge in China. Six years later the same authors (Liu & Guo 1988) proposed the combination *Mycovellosiella*

FIG. 1. *Passalora acericola*.

A, B. Leaf spots on *Acer pseudoplatanus*. C, Conidiophore. D. Conidium.

*acericola* (X.J. Liu & Y.L. Guo) X.J. Liu & Y.L. Guo, which Crous & Braun (2003) transferred to *Passalora* since *Mycovellosiella* was considered a synonym of that genus. This pathogen has also been reported from Italy on *Acer opalus* Mill. and from both Italy and Germany on *A. pseudoplatanus* (Braun & Crous 2005). This fourth record is the first report of *Passalora acericola* from Poland.

### Materials and methods

*Acer pseudoplatanus* (great maple, sycamore) is a native tree in Poland, often found in mountain and upland mixed forests. On lowlands, it is usually cultivated as an ornamental in parks and gardens and along roadsides. In Poland, *A. pseudoplatanus* reaches the northeastern limit of its natural range in Europe. The distinctive brownish lesions were collected from leaves of the tree in June 1989 and originally deposited in

the herbarium as *Cercospora acericola* Woron. After 20 years it was reexamined and redetermined as *Passalora acericola*.

The collected leaves of host plants were air-dried and examined by light microscopy (LM) in lactophenol Cotton Blue. The fungal nomenclature and taxonomy follows Crous & Braun (2003). The specimen examined is deposited in the herbarium of the Department of Botany and Mycology in Lublin (LBL M 8655).

### Taxonomy

*Passalora acericola* (X.J. Liu & Y.L. Guo) U. Braun & Crous,

*Mycosphaerella* and its anamorphs 1: 436. 2003.

FIGS. 1, 2

Leaf spots amphigenous, scattered, sometimes confluent, circular to subcircular, 1–4 mm in diameter, center grayish white, with wider yellowish brown halo and sometimes with border lines. Conidiophores solitary or 2–6 in fascicles, pale olivaceous-brown, straight or slightly curved, 0–1-septate, indistinct, conidial scars conspicuous, thickened and darkened,  $15\text{--}42.5 \times 4.5\text{--}6.5(-7)$

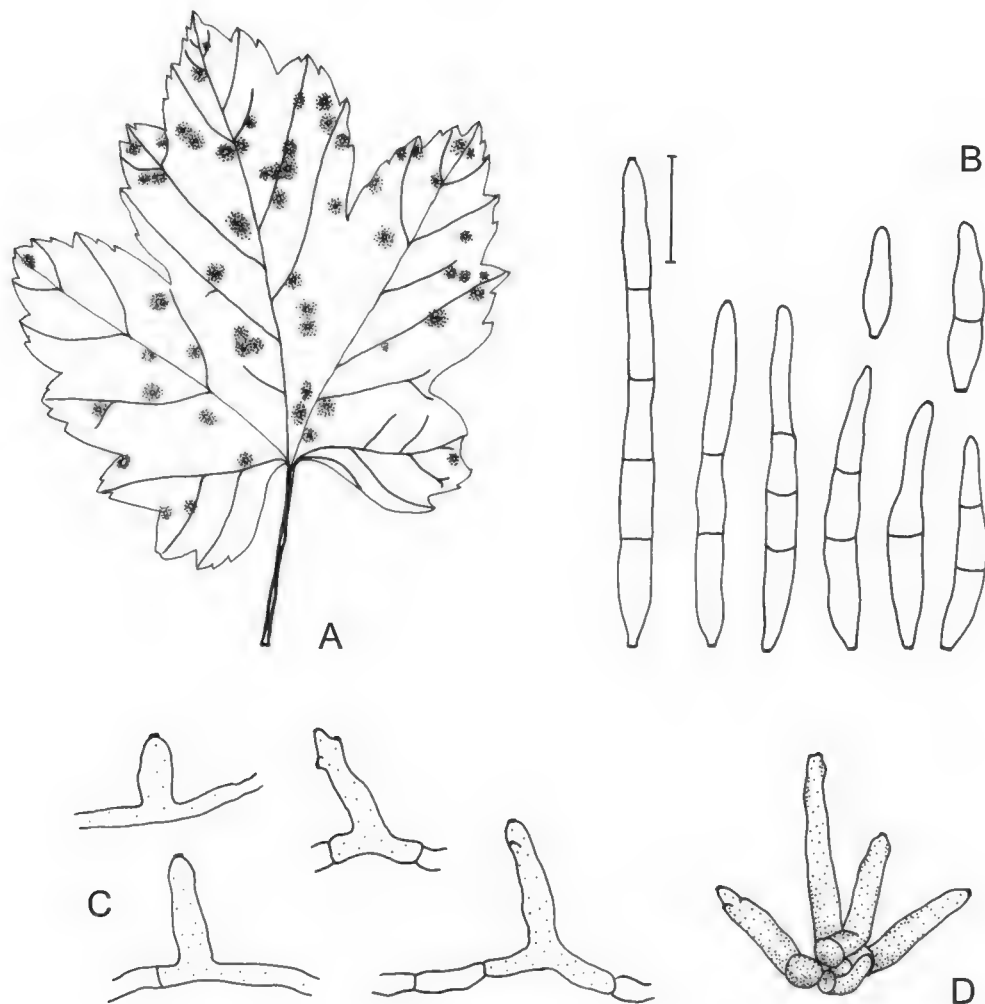


FIG. 2. *Passalora acericola* on *Acer pseudoplatanus*.

A. Leaf spots. B. Conidia. C, D. Conidiophores.

Scale bar = 20  $\mu\text{m}$ . U. Świdorska-Burek del.



µm. Conidia hyaline to subhyaline, solitary or occasionally catenate, obclavate to cylindrical, straight to slightly curved, usually 1–4-septate,  $35\text{--}85 \times 3.5\text{--}5$  µm, hila slightly thickened and darkened.

SPECIMEN EXAMINED: POLAND. WYŻYNA LUBELSKA UPLAND, Lipowiec village near Tyszowce town, on *Acer pseudoplatanus* L., 15 June 1989, W. Mułenko (LBL M 8655).

Several species of cercosporoid fungi have been reported worldwide on hosts of the genus *Acer*, including three species of *Cercospora* (*C. acerigena* U. Braun & Crous, *C. negundinis* Ellis & Everh., *C. saccharini* Liberta & Boewe), one species of *Pseudocercospora* (*Ps. acericola* (Woron.) Y.L. Guo & X.J. Liu), and only a single species of *Passalora* (*P. acericola*) (Crous & Braun 2003). The last species has previously been confused with *Pseudocercospora acericola* (= *Cercospora acericola* Woron.), which is, however, easily distinguishable by its inconspicuous, unthickened, non-pigmented loci (Braun & Crous 2005). The conidiophores in the Polish sample are somewhat wider than in the Chinese original description by Liu & Guo (1982), viz.  $15\text{--}42 \times 4.5\text{--}6.5(-7)$  (versus  $15\text{--}43.8 \times 3.8\text{--}6.3$  µm), but otherwise it agrees well with the type description.

### Acknowledgements

The authors would like to thank Uwe Braun (Halle/Saale, Germany) for his support in the identification of the pathogen and a presubmission review. We also thank Marcin Piątek (Kraków, Poland) for his valuable remarks.

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## MYCOTAXON

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**Occurrence of *Lentinula raphanica*  
in Amazonas State, Brazil**MARINA CAPELARI<sup>1</sup>, TATIANE ASAI<sup>1</sup> & NOEMIA KAZUE ISHIKAWA<sup>2</sup>*mcapelariibot@yahoo.com*<sup>1</sup> Instituto de Botânica, Núcleo de Pesquisa em Micologia  
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**Abstract** — *Lentinula raphanica* (Basidiomycota, Marasmiaceae) has been found in the Amazonian region of Brazil for the first time. Sequencing of the LSU region of the nuclear ribosomal DNA confirms the identity of the species. Macro- and microscopic descriptions and illustrations are provided, and the American distribution of *Lentinula* species is summarized.

**Key words** — Agaricales, diversity

**Introduction**

*Lentinula* Earle had long been considered to be a synonym of the cosmopolitan genus *Lentinus* Fr., but it is now accepted as a distinct genus with significant morphological differences in hyphal structure (Pegler 1983a) and type of wood rot (Redhead & Ginns 1985). Molecular data also confirm the distinction between the genera (Molina et al. 1992, Hibbett & Vilgalys 1993).

The genus comprises only seven morphological species with Asian-Australasian and American distributions. According to Nicholson et al. (1997), *Lentinula edodes* (Berk.) Pegler comprises three phylogenetic species with biological compatibility (Shimomura et al. 1992): *L. edodes*, *L. novae-zelandiae* (G. Stev.) Pegler, and *L. lateritia* (Berk.) Pegler. However, Fukuda et al. (1994) and Hibbett et al. (1998) have identified five distinct molecular groups within the *L. edodes* complex, each specific to a particular geographic region.

The currently recognized American species are *L. boryana* (Berk. & Mont.) Pegler, described from material collected by Blanchet de Laurane in Bahia State,

Brazil; *L. guarapiensis* (Speg.) Pegler, known only from the type collection made by Balansa in Guarapi, Paraguay; and *L. raphanica*, described from Florida, U.S.A, and segregated from specimens previously identified as *L. boryana* or *L. aciculospora* J.L. Mata & R.H. Petersen from Costa Rica.

*Lentinula boryana* and *L. raphanica* are morphologically very similar. Thon & Royse (1999) first established the separation of these two phylogenetic independent lineages within *L. boryana* after which Hibbett (2001) showed that the lineages represented two phylogenetic species of *Lentinula*. One species (“group VI”) had a Central American distribution, and the other (“group VII”) had a Coastal-Caribbean-South American distribution. Mata & Petersen (2001) and Mata et al. (2001) formally described these groups as *L. boryana* and *L. raphanica*, respectively.

In Brazil, *L. boryana* has been reported previously for Bahia, the type locality [Berkeley & Cooke 1876, as *Agaricus boryanus*; Dennis 1951, as *Collybia boryana*; Pegler 1983a; Mata & Petersen 2001], Paraná State (Meijer 2001), Rio Grande do Sul State (Rick 1907, as *C. boryana*; Singer 1952a, 1952b, 1953, as *Lentinus puiggarii*), and São Paulo (Grandi et al. 1984, as *Lentinus cubensis*; Pegler 1983b, under *L. puiggarii*; Pegler 1988, 1997). In this paper, we report the first record of *Lentinula raphanica* for Amazonas State, supported by morphological and molecular (nLSU) data. This is the second record for Brazil; the voucher material mentioned by Thon & Royse (1999, ambiguously cited as “sp834, Instituto de Botânica Herbarium, São Paulo, Brazil”) was not found there.

## Material and methods

### Sampling

The studied material was collected at the Instituto Nacional de Pesquisas da Amazônia (INPA), Manaus, Amazonas State. The specimens were deposited at the Instituto de Botânica Herbarium, São Paulo, Brazil (SP) and at the Instituto Nacional de Pesquisas da Amazônia Herbarium, Manaus, Brazil (INPA).

### Morphological study

Dried material was analyzed microscopically. Sections of basidiomata were first rehydrated with 70% ethanol and then with 5% KOH for 5–10 minutes and observed under a light microscope equipped with a drawing tube.

### Molecular study

nLSU rDNA sequences were phylogenetically analyzed to compare the *Lentinula* species from Amazonas, Brazil, with sequences deposited in GenBank (TABLE 1).

**DNA EXTRACTION** —DNA extraction protocols were adapted from Ferreira & Grattapaglia (1995) using lyophilized mycelium previously ground to a fine powder in liquid nitrogen. The sample was re-suspended in 50 µL TE, incubated at 37°C for 30 min after the addition of RNase A (0.01 mg µL<sup>-1</sup>), and stored at –20°C.

TABLE 1. Collection data and GenBank accession number of the taxa analyzed.

SPECIES	GENBANK NR.	VOUCHER/ STRAIN	REFERENCE
<i>Gymnopus biformis</i>	AF261336	RV98/32	Moncalvo et al. 2002
<i>G. menehune</i>	AY639423	AWW02-SFSU	Wilson & Desjardin 2005
<i>Lentinula boryana</i>	AF356151	R.G. Thorn 960624/09	Hibbett 2001
	AF356152	R38	Hibbett 2001
<i>L. lateritia</i>	AF356160	RHP3577	Hibbett 2001
	AF356162	TMI1172	Hibbett 2001
<i>L. raphanica</i>	AF356147	DUKE HN2002	Hibbett 2001
	GQ865600	SP394008	This study

PCR AMPLIFICATION — The 5’ end of the nLSU rDNA was targeted for amplification. The nLSU region was amplified using the primer set LR16 and LR5 (Moncalvo et al. 2000). PCR reactions containing 2.0 U of Platinum® Taq DNA Polymerase - Brazil (Invitrogen), 0.2 mM of each dNTP, 1.5 mM of MgCl<sub>2</sub> and 0.2 µM of each primer in 100 µL were performed in an Eppendorf thermocycler. The program was initiated by a 5-min denaturation step at 94°C, followed by 40 cycles of 40 sec at 94°C, 30 sec at 55°C and 60 sec at 72°C. Polymerization was completed by a 5-min incubation at 72°C. Amplification products were electrophoresed in a 1.5% agarose gel containing 0.1 µg ml<sup>-1</sup> ethidium bromide. PCR products were then purified using the PureLink PCR Purification Kit (Invitrogen).

DNA SEQUENCING —PCR product was sequenced in both directions using the same amplification primers and the DYEnamic ET Dye Terminator Kit in a MegaBACE 1000 DNA sequencer (GE Healthcare) according to the manufacturer’s instructions. The sequence was deposited in GenBank as GQ865600.

DATA ANALYSIS — Initially, a blast search was conducted in GenBank to compare the sequence obtained from the Amazonas material with existing sequence data. Subsequently, phylogenetic analysis was performed using the nLSU sequence determined in this study and five sequences available on GenBank (TABLE 1).

The sequences were analyzed using BioEdit version 7.0.5.3 (Hall 1999) and automatically aligned in Clustal W (Thompson et al. 1994). Parsimony analysis was performed using PAUP\* version 4.0b10 (Swofford 2001). The most parsimonious tree was obtained by a heuristic search with 1000 replicates of simple sequence addition, employing the tree-bisection-reconnection (TBR) branch-swapping algorithm. Characters from the extreme 5’ and 3’ ends of the sequences were deleted from all taxa to obtain individual datasets that had identical start and end positions. Gaps were treated as missing data, all characters were unordered and equally weighted, and multistate taxa were interpreted as uncertainty.

Branch support values were determined using 1000 bootstrap (BS) replicates. Estimated levels of homoplasy and phylogenetic signal (retention and consistency indexes) were determined. Trees were rooted using *Gymnopus biformis* (Peck) Halling and *G. menehune* Desjardin et al. as outgroups.

## Results and discussion

## Molecular analysis

Eight sequences were aligned — two each from three *Lentinula* taxa, and one per outgroup. The alignment consisted of 1426 characters, including gaps. Prior to analysis, 719 characters were excluded from the 5' and 3' ends of the sequences. Of 707 characters included in the analysis, 654 characters were constant, 10 variable characters were parsimony uninformative, and 43 were parsimony informative.

The heuristic search with 1000 BS replicates resulted in a single most parsimonious tree with the following scores: tree length = 60 steps, consistency index = 0.933, retention index = 0.934.

The most parsimonious tree generated from the nLSU sequence data from *Lentinula* species revealed three clades (FIG. 1) according with species identification. *L. boryana* is the sister species of *L. raphanica* with 75% BS support. Neighbor joining analysis (data not shown) showed the same topology. This result and a two-base pair difference between the Amazonas sequence (GQ865600) and the *L. raphanica* sequence (AF356147, obtained from the same material (DUKE HN2002) used for ITS analysis by Mata et al. 2001) support the Amazonas material as *L. raphanica*.

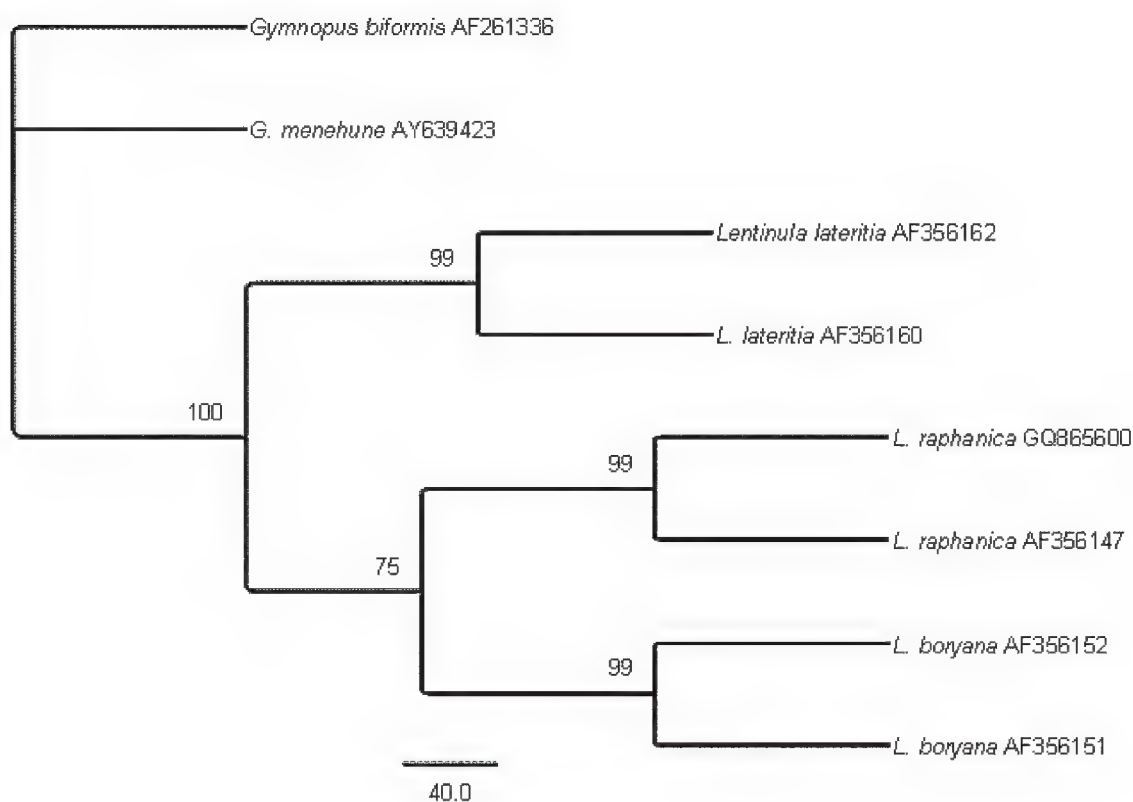


FIGURE 1. MP tree generated by parsimony analysis of partial LSU rDNA sequences. BS values are shown above branches. GenBank accession numbers are shown after each taxon name.



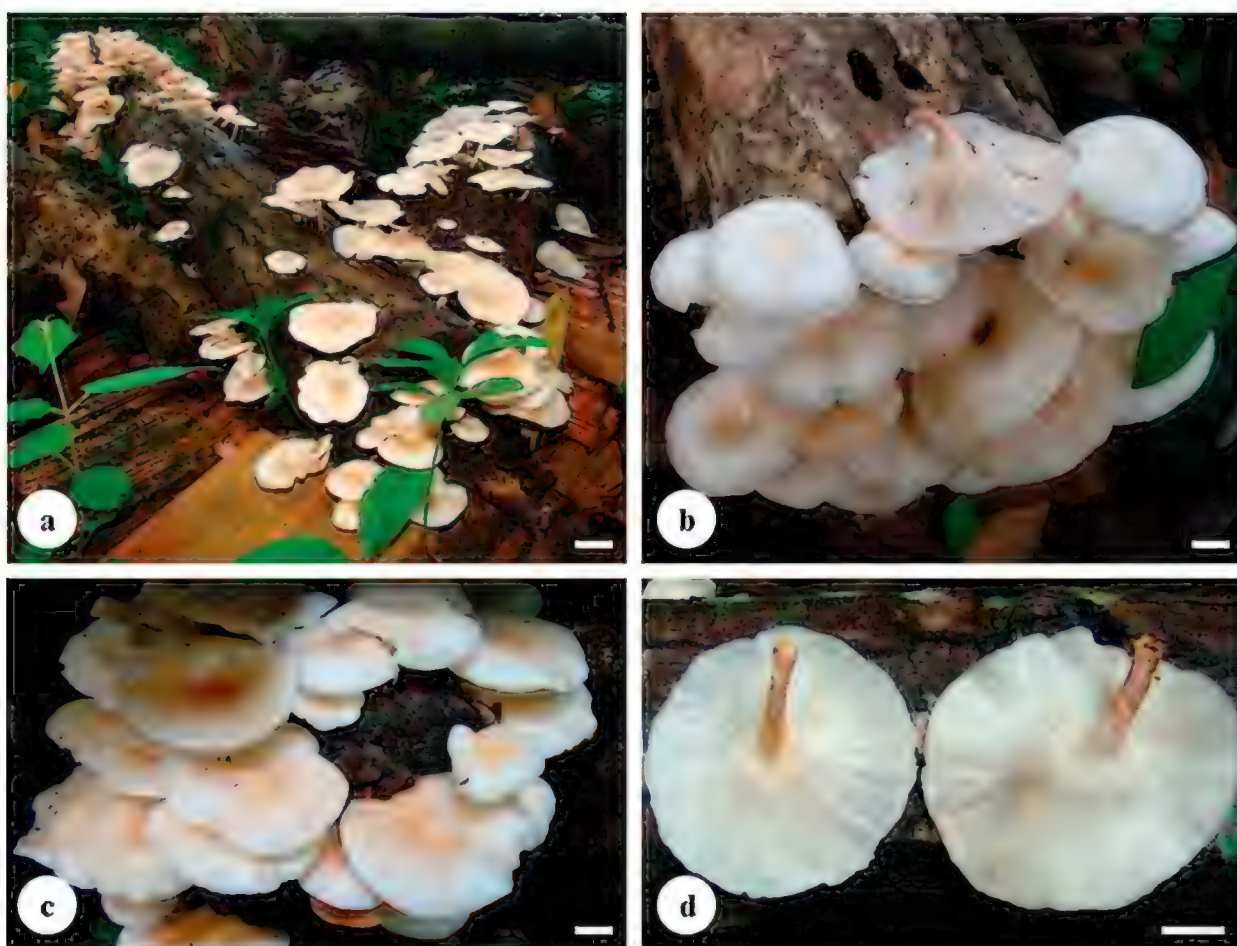


FIGURE 2. *Lentinula raphanica* (INPA230870, SP394008). Bar = 1 cm.

## Taxonomy

*Lentinula raphanica* (Murrill) J.L. Mata & R.H. Petersen, Mycotaxon

79: 228. 2001.

FIGS. 2, 3

≡ *Armillaria raphanica* Murrill, Mycologia 35: 423. 1943.

≡ *Lentinula raphanica* (Murrill) J.L. Mata & R.H. Petersen,  
Mycologia 93: 1107. 2001 (superfluous combination).

= *Gymnopus alliaceus* Murrill, Mycologia 35: 425. 1943.

PILEUS 3–6 cm diam., convex at first with an involute margin, then applanate with a depressed center to infundibuliform when fully expanded, glabrous, some slightly viscous, smooth, hygrophanous, white to dirty white, center sometimes cinnamon brown to brownish, sometimes with cinnamon brown patches, fleshy. LAMELLAE free, crowded, white, thin, smooth-edged, with lamellulae. STIPE 40 × 3 mm, central to slightly eccentric, curved, equal to tapering at the base, surface with some floccose fibrillose small scales, dirty white to pinkish, with brownish base, firm. Annulus absent. BASIDIOSPORES not seen. BASIDIA not seen. BASIDIOLES numerous, mostly ventricose, 13–18 × 4–5 µm. PLEUROCYSTIDIA absent. CHEILOCYSTIDIA 21.4–28.5 × 5–7 µm, versiform, contorted, clavate with diverticulate outgrowths, hyaline, thin-walled, clamped at the base. LAMELLAR TRAMA regular, becoming interwoven towards the edge,

hyphae 3–14  $\mu\text{m}$ , hyaline, thin to slightly thick-walled, with clamp connections. CAULOCYSTIDIA 14.2–50  $\times$  2–7  $\mu\text{m}$ , abundant, cylindrical, clavate or flexuous, apex obtuse or with outgrowths, hyaline. Lignicolous, growing on *Bertholletia excelsa* Humb. & Bonpl. (*Lecythidaceae*, castanha da amazônia).

EXAMINED MATERIAL — BRAZIL. AMAZONAS: Manaus, INSTITUTO NACIONAL DE PESQUISAS DA AMAZÔNIA — 04.XII.2007, N.K. Ishikawa s.n. (INPA230870, SP394008; GENBANK GQ865600); 31.VIII.2007, T.A. Silva s.n. (INPA230868, SP394011); 21.XI.2007, T.A. Silva s.n. (INPA230869, SP394010).

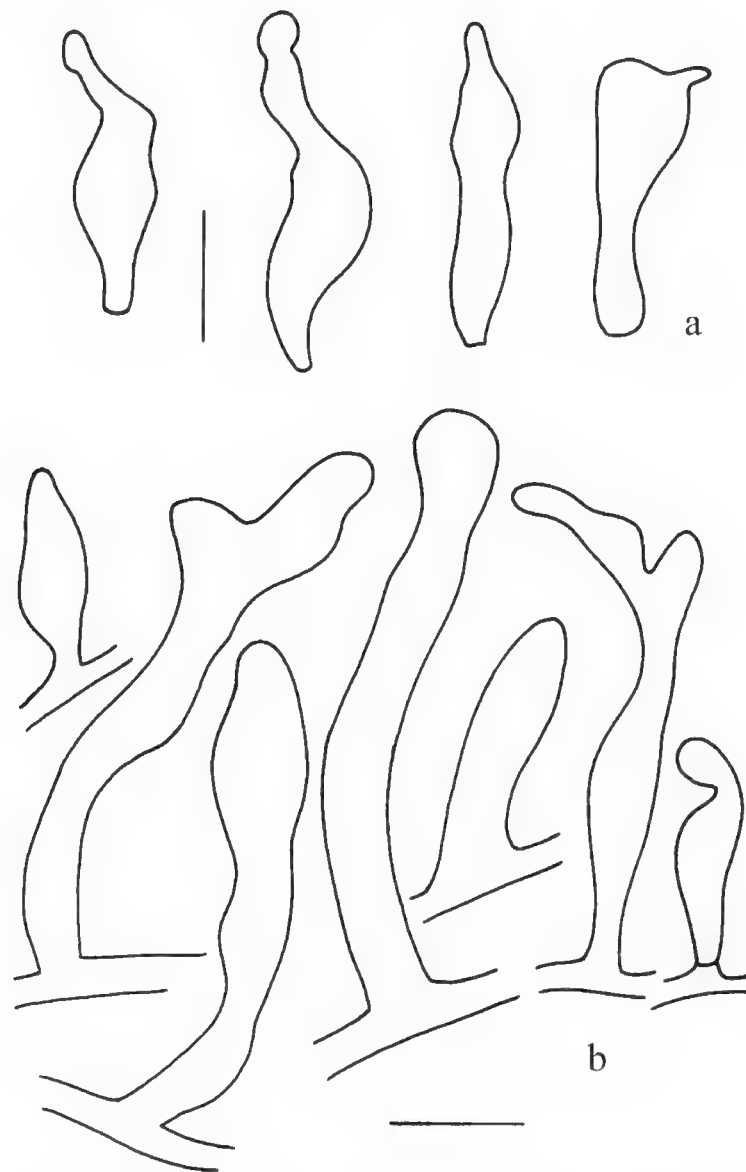


FIGURE 3. *Lentinula raphanica*. a. Cheilocystidia. b. Caulocystidia. (INPA230869, SP394010). Bar = 10  $\mu\text{m}$ .

Except for the lighter pileus colour, the Amazonas specimens fit very well macroscopically with the description by Mata et al. (2001). *Lentinula raphanica* greatly resembles *L. boryana*, differing mainly in the cheilocystidia shape and basidiospore dimensions (Mata et al. 2001). According to Mata et al. (2001), in *L. raphanica* basidiospores “have a narrower shape, more subcylindrical than oblong”, measuring 4.8–7.2  $\times$  2.0–3.6  $\mu\text{m}$ ,  $Q = 1.50\text{--}3.00$ ,  $Q_x = 2.16$ ,

while in *L. boryana* they measure  $4.8\text{--}8.0 \times 2.4\text{--}4.0 \text{ }\mu\text{m}$ ,  $Q = 1.30\text{--}2.67$ ,  $Q_x = 1.91$ . Unfortunately, the three Amazonas collections examined were sterile, lacking basidia on the lamellae, and the cheilocystidia were very difficult to see. Nevertheless, it was possible to confirm the species identity by matching the caulocystidia shape to those depicted by Mata et al. (2001). In *L. boryana*, the caulocystidia are cylindrical to clavate, while in *L. raphanica*, they are cylindrical, clavate, or flexuous, with an obtuse apex, and knobbed or with outgrowths. This difference in shape seems to be a constant and reliable diagnostic character. A more complete description of this species can be found in Mata et al. (2001) and Mata & Petersen (2001).



FIGURE 4. Geographic distribution of the American species of *Lentinula*.

The geographic distribution of the American species of *Lentinula* is shown in FIG. 4. After Mata & Petersen (2000) and Mata et al. (2001), *L. raphanica* was known from Brazil (probably from São Paulo State), Costa Rica, Puerto Rico, Trinidad, United States of America, and Venezuela; *L. boryana* was known from Brazil, Costa Rica, Cuba, Guadeloupe, Guyana, Mexico and Panama; *L. aciculospora* was known from Costa Rica; and *L. guarapiensis* was known from Paraguay. Subsequently, Vasco-Palacios et al. (2005) have reported *L. raphanica* from Colombia and Piepenbring (2008) has recorded *L. aciculospora* in Panama. Except for *L. guarapiensis*, which should be re-collected at or near its type locality to establish its biological and phylogenetic identity, the remaining species are well defined and probably occur throughout Central and South America. Further explorations will add *Lentinula* collections and may improve understanding of its distribution and diversity in the Americas.

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## MYCOTAXON

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***Pseudocercospora heliconiae* sp. nov. causing leaf blight on parakeet flower, *Heliconia psittacorum* (Heliconiaceae), in Brazil**

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**Abstract** — The leaf spotting hyphomycete *Pseudocercospora heliconiae* sp. nov., collected on *Heliconia psittacorum* in a commercial nursery in Viçosa, Minas Gerais State, Brazil, is described, illustrated, discussed and compared with allied species.

**Key words** — cercosporoid, ornamental plant, phytopathology, plant disease, taxonomy, tropical fungi

**Introduction**

In May 2008, a severe leaf spot disease was observed on *Heliconia psittacorum* L. f. (parakeet flower) in Minas Gerais State, Brazil. A fungus belonging to the genus *Pseudocercospora* Speg., was consistently found associated with the symptoms observed. This important ornamental species is known as an alternative host for *Pseudocercospora fijiensis* (M. Morelet) Deighton (the agent of the black leaf streak of banana) in Brazil, the sole alternative host not belonging to the genus *Musa* L. Morphological studies and pathogenicity tests were conducted to elucidate the disease aetiology. The fungus was proved to be distinct of *P. fijiensis* and other related *Pseudocercospora* spp. on *Musaceae* and proposed as a new species within the genus *Pseudocercospora*. This new species is described, illustrated, and discussed in this paper.

**Material and methods**

Samples of *H. psittacorum* infected with *P. heliconiae* were collected, photographed (SONY DSC-H9 digital camera), dried in a plant press and deposited at the herbaria VIC and HAL. Under a stereomicroscope, selected structures of the fungus were removed from fresh leaf spots and mounted in glass slides with lactophenol. Observations, measurements and illustrations were carried out by

means of an OLYMPUS BX 50 light microscope fitted with a digital camera (EVOLT E330) and a drawing tube. Wherever possible, 30 measurements were made of the structures mounted. To perform the pathogenicity tests, the fungus was isolated onto PDA, brought into pure culture and grown at 27°C for 20 days. Cultures disks were taken from the border of the colonies and used to inoculate four healthy young and mature leaves of *H. psittacorum* and banana plants (cv. Prata Anã). The inoculated plants were maintained in moist chambers for two days and then transferred to a greenhouse at 25°C. Leaves of both species, on which only PDA plugs were placed, served as control.

### Taxonomic description

*Pseudocercospora heliconiae* Meiriele Silva & O.L. Pereira, sp. nov.

FIGS 1–6

MYCOBANK 518119

*Maculae amphigenae, irregulares, necroticae, brunneae, confluentes. Stromata nulla vel minuta. Caespituli saepe hypophylli, atro-brunnei. Conidiophora laxae vel dense fasciculata, pauca vel modice numerosa, per stoma emergentia, recta vel curvata, cylindrica, non ramosa, 22.5–77.5 × 5.0–8.75 µm, medio-brunnea, laevia, 0–2 septata. Cellulae conidiogenae integratae, terminales, laeviae, cicatrices conidiales inconspicuae. Conidia, solitaria, pallide brunnea, cylindrica, recta ad leviter curvata, 52.5–120.5 × 4.5–6.0 µm, apice obtuso, basi truncata, hila non incrassata, non fuscata, 0–5-septata, laevia.*

**HOLOTYPE:** BRAZIL, Minas Gerais, Viçosa, on leaves of *Heliconia psittacorum* L. f. (*Heliconiaceae*), 12 May 2008, O. L. Pereira (VIC 31221). **Isotype:** HAL 2356 F.

**ETYMOLOGY:** referring to the host genus *Heliconia*.

Leaf spots amphigenous, irregular, necrotic, brownish, confluent, covering large areas of the leaf surfaces. Stromata absent or small. Caespituli mainly hypophyllous, dark brown. Conidiophores in loose to dense, small to moderately large fascicles, straight to curved, cylindrical, unbranched, 22.5–77.5 × 5.5–8.75 µm, 0–2 septate, medium brown, smooth. Conidiogenous cells integrated, terminal, smooth, scars inconspicuous. Conidia solitary cylindrical, straight to slightly curved, 52.5–120.5 × 4.5–6.0 µm, 0–5-septate, pale brown, smooth, apex obtuse, base truncate, hilum neither thickened nor darkened.

**COMMENTS** — Necrotic symptoms, similar to those originally observed in the field, were detected 10 days after inoculation only on mature leaves of *H. psittacorum*. Inoculated leaves of cv. Prata Anã and uninoculated control leaves, on which only PDA plugs were placed, remained healthy. The fungus was then reisolated, satisfying Koch's Postulates.

Only two cercosporoid fungi are known to occur on members of the genus *Heliconia* L., viz. *Cercospora heliconiae* Chowdhry et al. reported on *Heliconia caribaea* Lam. in India (Crous & Braun 2003) and *Pseudocercospora fijiensis* reported on *H. psittacorum* in Brazil (Gasparotto et al. 2005). *Cercospora heliconiae* is considered to be a true *Cercospora* s. str., close or identical to



FIGS 1–2. *Pseudocercospora heliconiae*. 1. Leaf blight on *Heliconia psittacorum* from a commercial nursery for cut flower in Viçosa, Minas Gerais, Brazil. 2. Detail of coalescent lesions on leaves.

*Cercospora apii* Fresen. s. lat. (Crous & Braun 2003). *Pseudocercospora fijiensis*, the causal agent of the black leaf streak of banana, is the sole *Pseudocercospora* reported on the genus *Heliconia*. However, *P. fijiensis* has very diagnostic scars and hila (*Paracercospora*-like, thickened and darkened ultimate rim) (Mulder & Holliday 1974), which were not observed in the samples of *H. psittacorum* from Minas Gerais. Additionally, despite the conidia of *Pseudocercospora heliconiae* resemble those of *P. fijiensis* in color, they are wider and longer.

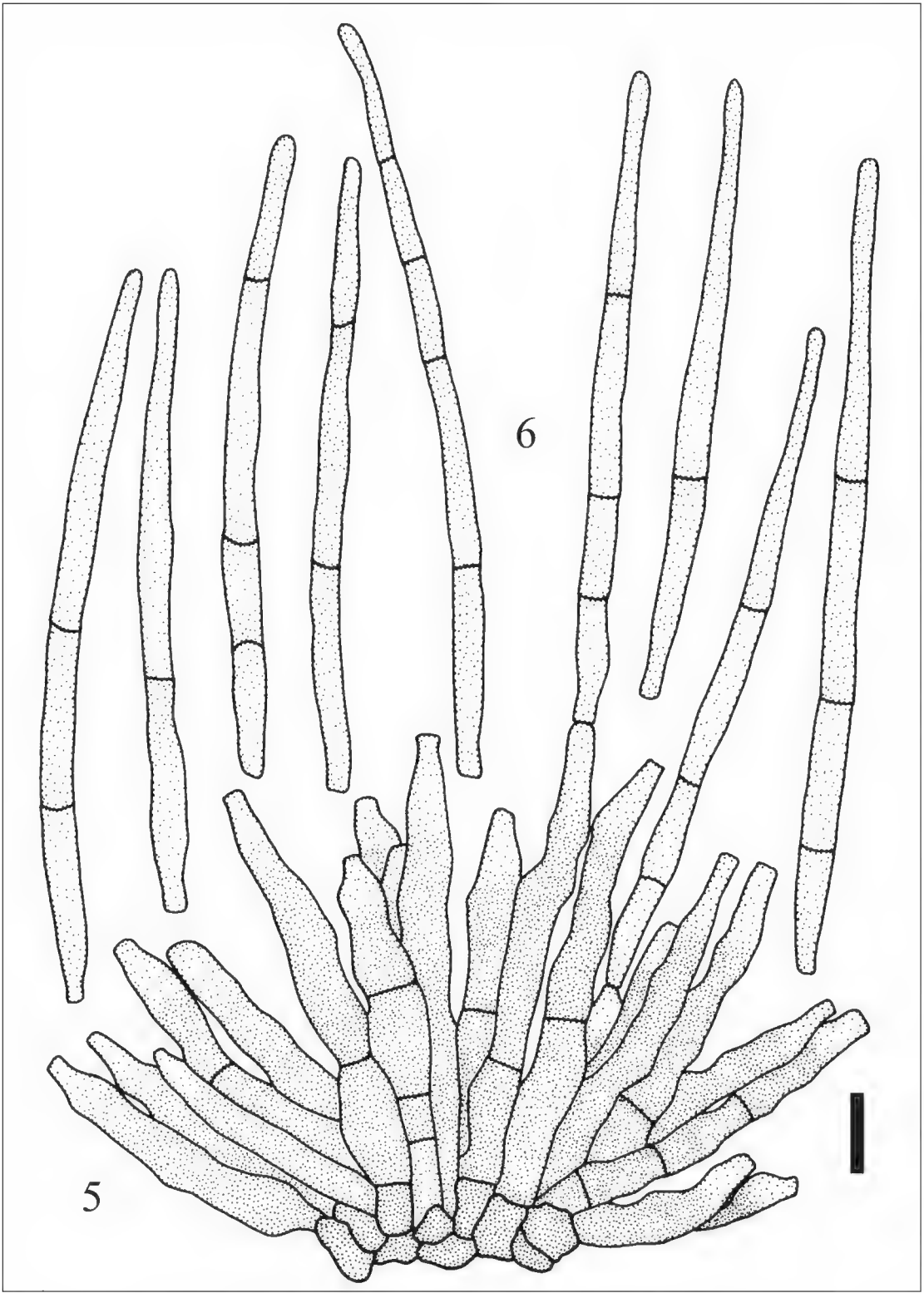
As *Heliconiaceae* was previously regarded a subfamily within *Musaceae* we compared *P. heliconiae* with nine additional *Pseudocercospora* spp. that have been recorded on *Musaceae*. *Pseudocercospora assamensis* Arzanlou & Crous, *P. indonesiana* Arzanlou & Crous, *P. musae-sapientum* (A.K. Kar & M. Mandal) U. Braun & Mouch., *P. fengshanensis* (T.Y. Lin & J.M. Yen) J.M. Yen & S.K. Sun, *P. musicola* U. Braun, *P. vanieriae* (Chupp & Linder) U. Braun & Crous, *P. musae* (Zimm.) Deighton and *P. eumusae* Crous & Mour., can be distinguished of *P. heliconiae* by having shorter conidia (Chupp 1954, Hsieh & Goh 1990,





FIGS 3–4. *Pseudocercospora heliconiae*. 3. Leaf blight on a severely infected plant leading to leaf death. 4. Detail of blight symptom covering the whole leaf surface.





FIGS 5–6. *Pseudocercospora heliconiae* (VIC 31221, holotype).  
5. Fasciculate conidiophores with inconspicuous conidiogenous cells.  
6. Cylindrical conidia with truncate inconspicuous hila.  
Scale bar: 10  $\mu$ m.

Braun et al. 1999, Crous & Mourichon 2002, Arzanlou et al. 2008), while *P. longispora* Arzanlou & Crous has narrower conidia (Arzanlou et al. 2008). Hence, the introduction of a new species is undoubtedly justified.

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## MYCOTAXON

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**First record of *Tulostoma gracilipes*  
(Agaricales, Agaricaceae) for the Americas**CAROLINA PIÑA<sup>1</sup>, MARTÍN ESQUEDA<sup>1\*</sup>,  
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**Abstract** — *Tulostoma gracilipes* is reported for the first time in the Americas. Since this species was only known in the type locality of South Africa, this record from Mexico represents the second worldwide. Observations of macro- and microscopic characters for the holotype and Sonoran collection are presented. SEM photomicrographs illustrating spore ornamentation are included.

**Key words** — *Agaricomycetes*, gasteromycetes, chorology, taxonomy

**Introduction**

Wright (1987) included 138 taxa in the world monograph of *Tulostoma*. Some species such as *T. gracilipes* and *T. portoricense* J.E. Wright were only known in the type locality given in the monograph. *Tulostoma portoricense* was reported for the second time worldwide (Esqueda et al. 1998), from the mycobiota of Sonora, Mexico. Here we report the first record of *T. gracilipes* from North America, also from Sonora; previously, it was known only from Africa.

Twenty-seven taxa of *Tulostoma* have been registered in Sonora, Mexico (Esqueda et al. 2010). Some of them are broadly distributed: *T. fimbriatum* Fr., *T. squamosum* (J.F. Gmel.) Pers., and *T. pulchellum* Sacc. (Esqueda et al. 2004). Other species have a restricted distribution: *T. floridanum* Lloyd, *T. submembranaceum* G. Moreno et al., and *T. mohavei* Lloyd. This last species was found in the Pinacate and Great Altar Desert Biosphere Reserve (Esqueda et al. 2006).

*Tulostoma gracilipes* was collected in a protected natural area of Sonora: the Sierra de Mazatán, which is located in central region of Sonora and belongs to the Sonoran Desert Province (28°58'–29°30'N, 109°59'–110°33'W; INEGI 2009). According to the Commission for the Knowledge and Use of Biodiversity in Mexico (CONABIO), this area is an “island” of temperate biodiversity surrounded by the arid landscape of the Sonoran Desert (Arriaga 2000). The predominant vegetation type is subtropical scrub, with oak forest in the highest areas, and semiarid plains with mesquite scrub. This is the first report of a fungus for the Sierra de Mazatán.

### Materials and methods

The specimen has been deposited in the macromycetes collection of the Centro de Estudios Superiores del Estado de Sonora (CESUES). Observations of microscopic characters (e.g., spore dimension, including ornamentation) were made using a light microscope to observe material mounted in Hoyer's medium. For ultrastructural studies (e.g., spore ornamentation characteristics), the sample was prepared according to the critical-point-drying method outlined in Moreno et al. (1995) and examined with a Zeiss DSM-950 scanning electron microscope.

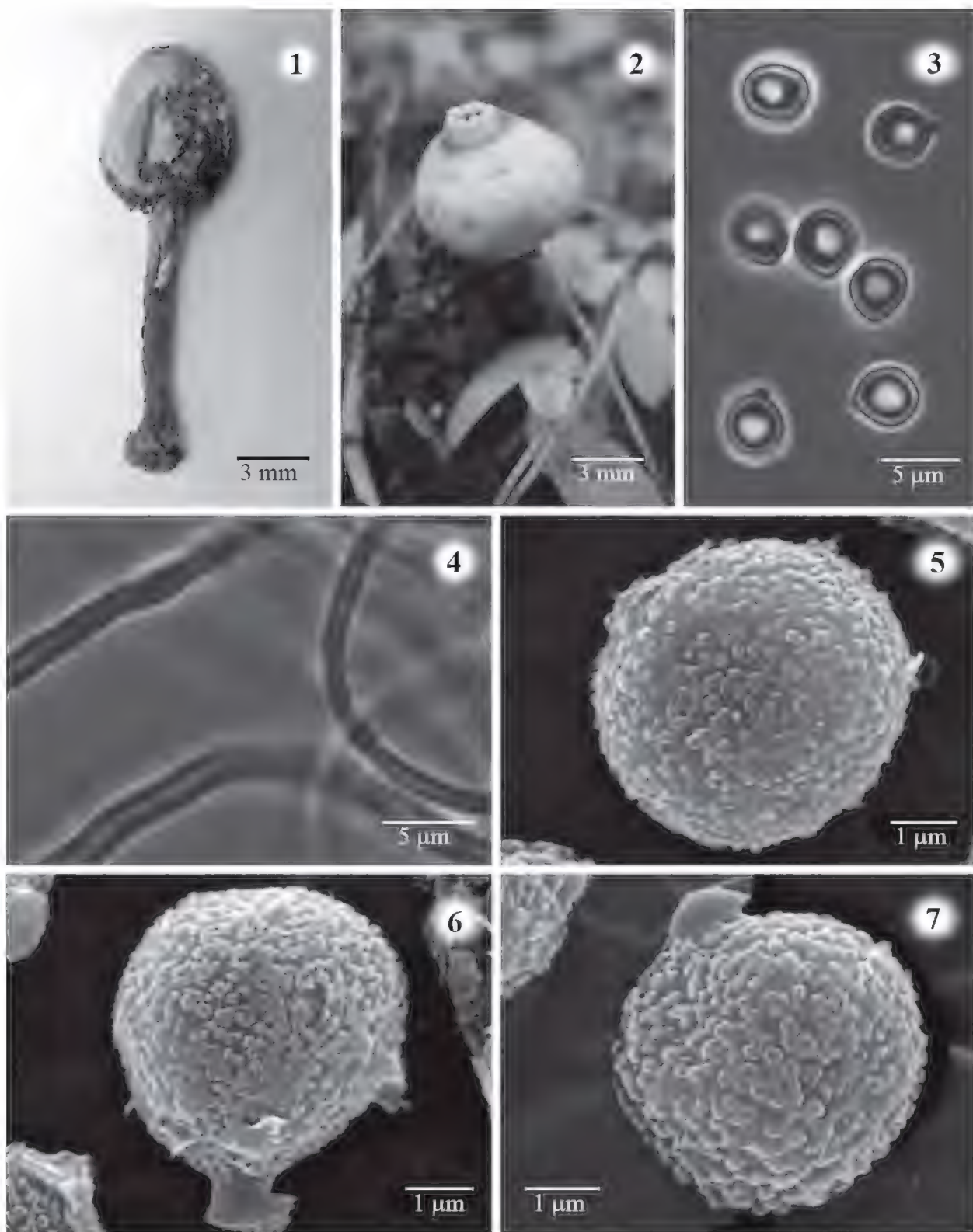
### Taxonomy

*Tulostoma gracilipes* J.E. Wright, *Bibliotheca Mycol.* 113: 125 (1987)

SPECIMENS EXAMINED — MÉXICO. SONORA: Municipality of Ures, leg. C. Piña & A. Gutierrez, 18.XII.2008, CESUES 9100. SOUTH AFRICA. NORTHERN CAPE: Lokenburg, leg. J.P.H. Acocks 18.934, 18.VIII.1956, PREM 41.614, Holotype.

Spore sac  $8 \times 7$  mm. Exoperidium membranous, cream coloured within and dirty cream outside covered by soil grains, persistent mainly at spore sac base (FIG. 1). Endoperidium glabrous, little velvet surface under stereoscopic microscope, white isabelline. Stoma fibrillose-fimbriate, opening less than 1 mm diam., scarcely projecting lip ca. 0.5 mm wide, with a denticulate aspect, surrounded by an easily seen brownish spore deposit simulating a coloured peristome (FIG. 2). Socket shallow, membranous with irregular margins, close to stem. Gleba ochraceous. Stem gracile, yellowish to light brown, longitudinally substriate,  $14 \times 2$  mm, ending basally in a mycelial bulb (FIG. 1).

Spores  $3.6\text{--}4.9 \times 3.1\text{--}4.6$   $\mu\text{m}$ , yellowish, smooth under LM (Fig. 3), globose, subglobose to ellipsoid, guttulate, thick-walled, hilar appendage  $0.5 \times 1$   $\mu\text{m}$ . Capillitium (Fig. 4) of  $1.7\text{--}3.8$   $\mu\text{m}$  diam.,  $0.5\text{--}1.5$   $\mu\text{m}$  thick wall, lumen visible to solid, flexuose, scantily septate and branched, wall conspicuously encrusted with inorganic matter particles. Under SEM spores are verrucose, with small and densely crowded verrucae (FIGS. 5–7) that are occasionally joined.



FIGS. 1–7. *Tulostoma gracilipes* (CESUES 9100):  
 1. Basidiome. 2. Spore sac detail. 3. Spores under LM. 4. Capillitium under LM.  
 5–7. Spores under SEM.

HABITAT — Sandy soils, mesquite vegetation, under *Phaulothamnus spinescens* A. Gray (*Achatocarpaceae*) among litter, during autumn.

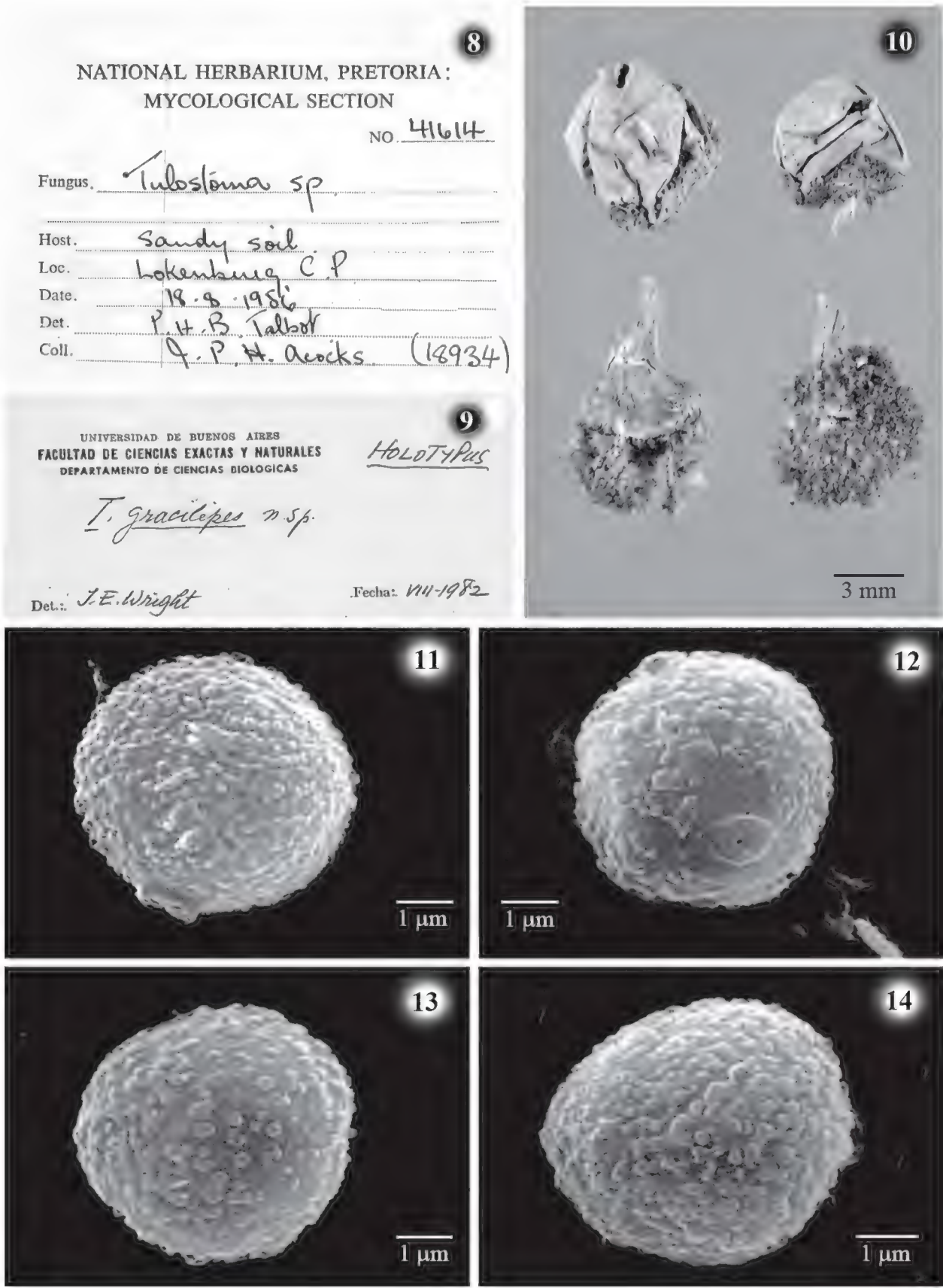


OBSERVATIONS — The Sonoran material was compared with the type collection of *T. gracilipes* kept in Pretoria (PREM) (FIGS. 8–9), which allowed us to confirm our determination. The two basidiomes examined in that type (FIG. 10) had very fragmented stems, but their bases are in good condition. According to the remains, the stems were slender, ca. 1 mm diam., but it was impossible to determine length. In his original description, Wright (1987) describes the stems as up to 25 mm long, which fits well with the illustration included in the monograph (pl. XLIV: 5). He also described the stems as “gracile”, a feature reflected in the name of the species. A notable bulbiform thickening (5.5 mm) is also observed at the base of one stem in the type. Stipe bases of both basidiomes are covered by fragile mycelial remains that agglomerate sand grains.

The spore sacs are small (6–7 mm diam.) and well preserved. The endoperidial surface is slightly velvety. Other remarkable features in the type collection are those of the stoma and exoperidium. The stoma is fibrillose-fimbriate, slightly projecting as a denticulate lip, concolorous. The exoperidium persisting on the base of spore sacs is typically membranous, and externally covered by a hyphal layer mixed with sand grains. Spores 4–5.5  $\mu\text{m}$  diam., globose to irregular, yellowish, subsmooth to slightly asperulate under LM. Under SEM spore ornamentation consists of numerous low verrucae, sometimes slightly flattened (FIGS. 11–14). Capillitium 4–13  $\mu\text{m}$  diam., uncoloured, thick walled up to 2–3  $\mu\text{m}$ , leaving an irregular lumen, and with scarce uncoloured septa due to their disarticulation.

*Tulostoma parvissimum* Long & S. Ahmad is closely related but shows an almost indefinite fibrillose stoma and asperulate spores (LM) with larger verrucae. The similar *T. berteroanum* (Lév.) Sacc. has a mouth that is mammoscutesellate to fibrillose when mature and basidiospores with conspicuously larger verrucae under SEM. With small basidiomes and almost identical spores even under the SEM, *T. herteri* Lohwag & Swoboda is a very similar species, which, however, differs notably from *T. gracilipes* in its mammoscutesellate stoma and hyphal exoperidium (Dios et al. 2004). We could also consider the similarity of the spores of *T. pulchellum* (which also has a membranous exoperidium), but its verrucae are usually more flattened and sometimes united in short crests. Besides, *T. pulchellum* shows mammoscutesellate stoma and more robust basidiomes.

In conclusion, *T. gracilipes* is easily recognized by the combination of the following characters: fibrillose-fimbriate to denticulate stoma, membranous exoperidium, spores that are subsmooth under LM and verrucose under SEM, and a basidiome that is minute. This second world record of *T. gracilipes* allows us to extend significantly the distribution area of the species.



FIGS. 8–14. Holotype of *Tulostoma gracilipes*:  
8–9. Labels. 10. Basidiome. 11–14. Spores under SEM.

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We wish to express our gratitude to Dr. Hanns Kreisel and Dra. María Martha Dios for reviewing the manuscript and offering useful comments. Thanks to M. en C. Felipe Barredo-Pool of the Electron Microscopy Service of the Centro de Investigación Científica de Yucatán A.C. for their invaluable help with the SEM and to Bianca Delfosse for revising the English.

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## MYCOTAXON

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***Chondrogaster pachysporus* in a  
*Eucalyptus* plantation of southern Brazil**

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**Abstract** — *Chondrogaster pachysporus* is reported for the first time in Brazil. It is similar to *C. angustisporus*, also known from southern Brazil, but differs in the size and ornamentation of the basidiospores and in the presence of monosporic basidia. The hypogeous sequestrate specimens were collected in a *Eucalyptus saligna* plantation. Descriptions, photographs, and line drawings of the specimens are presented.

**Key words** — ectomycorrhiza, false-truffle, *Hysterangiales*, *Mesophelliaceae*

## Introduction

*Chondrogaster* Maire is a genus of sequestrate fungi characterized by enclosed hypogeous basidiomata that bear a loculate gleba composed of tramal plates where basidia and basidiospores are produced (Castellano et al. 1989). The genus is closely related to *Hysterangium* Vittad., from which it was segregated and differs in the lack of a distinct columella and presence of a mycelial mass covering the whole basidioma (Giachini et al. 2000). Both currently known species are associated with *Eucalyptus* and possibly native to Australia but have spread to many areas where *Eucalyptus* plantations have been established for forestry purposes.

*Chondrogaster angustisporus* Giachini et al., originally described from Australia, Uruguay, and southern Brazil (Giachini et al. 2000), is possibly the only South American record of the genus. *Chondrogaster pachysporus*, the type

species, is so far known from the Mediterranean zone (Europe and Africa), North America, and Australia (Lago & Castro 2004). Recent studies on the biology and taxonomy of sequestrate fungi in Brazil and neighboring countries are scarce and limited to a few local revisions from Brazil (Giachini et al. 2000, Cortez et al. 2008) and Argentina (Nouhra et al. 2005, 2008). In this paper, we report the occurrence of *C. pachysporus* in southern Brazil.

## Materials and methods

Fieldwork was conducted in a *Eucalyptus saligna* Sm. plantation at the Experimental Forestry Station (FEPAGRO), in the municipality of Santa Maria, central region of Rio Grande do Sul State, southern Brazil (29°45' S, 53°43'W). The site comprises 280 ha for cultivation of native trees as *Apuleia leiocarpa* (Caesalpinaceae), *Senna multijuga* (Caesalpinaceae), and *Tabebuia* spp. (Bignoniaceae), and exotics as *Hovenia dulcis* (Rhamnaceae), *Platanus acerifolia* (Platanaceae), *Pinus* (Pinaceae), and *Eucalyptus* spp. (Myrtaceae). Soil is of the Hapludult type, which is deep, imperfectly drained and with low natural fertility (Abrão et al. 1988, Streck et al. 2008). Climate is subtropical humid (Cfa) according to Köppen's system, with mean temperature values for the warmest month higher than 22°C (Menegat 1998). Annual rainfall is about 1769 mm, with rains well distributed throughout the year (Schumacher et al. 2008).

Fresh basidiomata were collected and photographed in situ, then analyzed macro- and microscopically following Brundrett et al. (1996) and Castellano et al. (2004). Color names and codes follow Kornerup & Wanscher (1978). Microscopic analysis of the basidiomata comprised 30 measurements of each microstructure (basidiospores, basidia, and hyphae), which were drawn under a light camera. Specimens are deposited in the herbaria of Department of Biology, "Universidade Federal de Santa Maria" (SMDB) and the Institute of Biosciences, "Universidade Federal do Rio Grande do Sul" (ICN).

## Taxonomy

*Chondrogaster pachysporus* Maire, Bull. Soc. Mycol. Fr. 40: 312, 1925. FIG. 1–8

BASIDIOMATA hypogeous, 8–23 mm in width, 7–11 mm high, depressed globose to subglobose, aggregated in clusters within a common mycelium. PERIDIUM <1 mm thick, greyish beige (4C2) when fresh, dull red (9C4) when bruised, glabrous or covered by scattered to numerous rhizomorphs. GLEBA composed by non-gelatinized, radially arranged locules, greyish green (28C3) to dull green (29D3) at younger stages, to finally olive (1F5) or blackish at maturity. RHIZOMORPHS white, numerous, arising from several points of attachment in the basidiomata surface. COLUMELLA absent.

BASIDIOSPORES 12.5–16.5 × 6–9 µm (ornamentation excluded), subfusoid, ellipsoid to broad ellipsoid, apex and base tapered, some with a shortly mucronate apex; sterigmal attachment persistent at maturity; in KOH, they are hyaline when young to finally pale yellowish brown at maturity; wall smooth



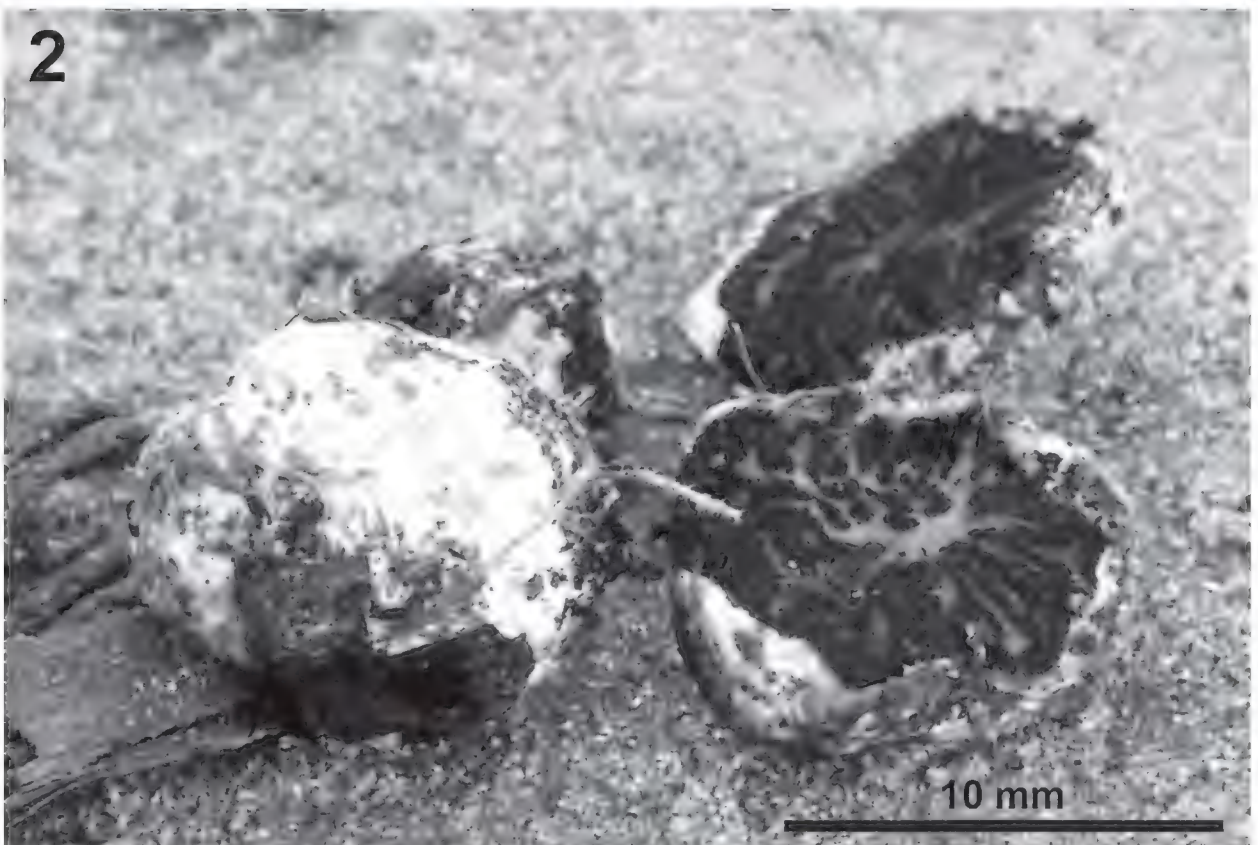
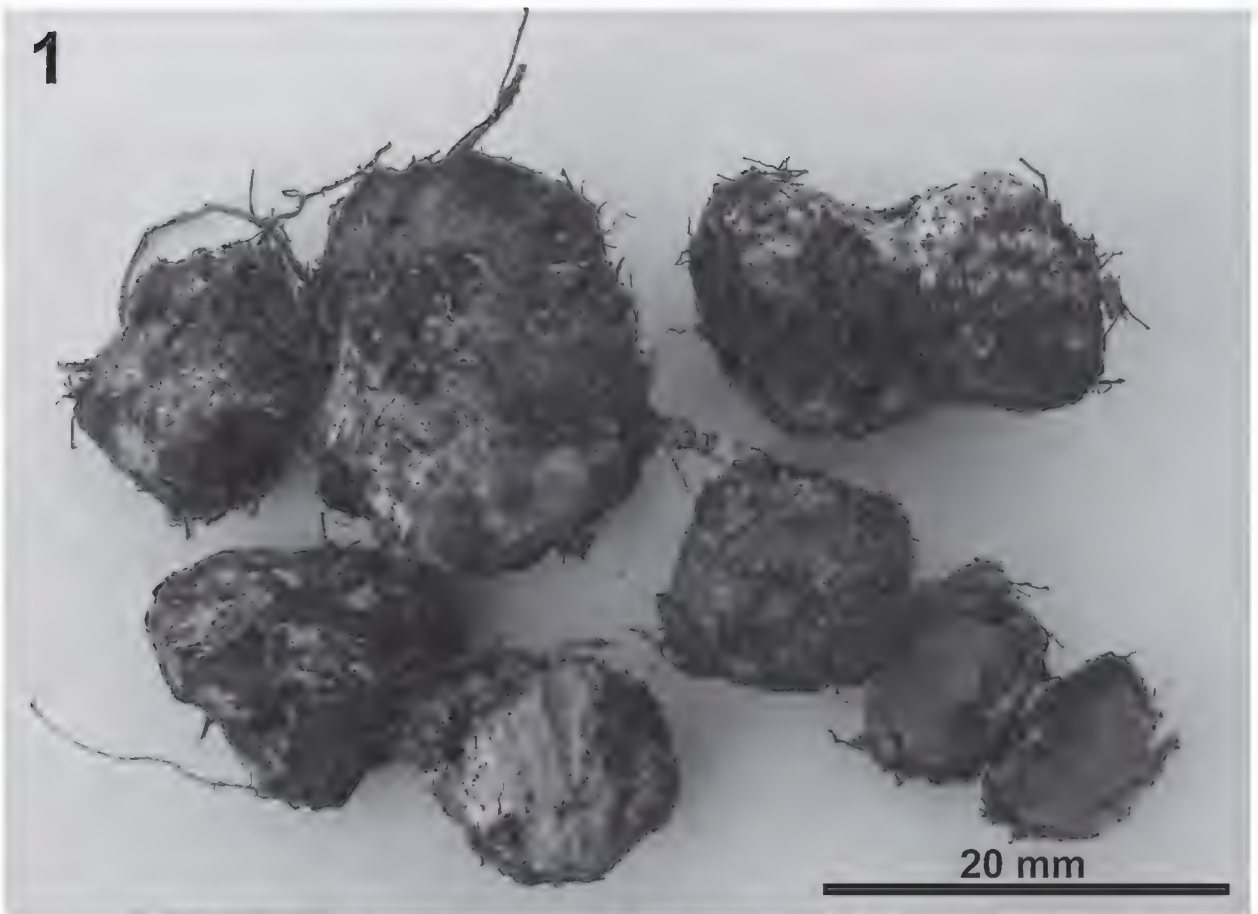


FIG. 1-2. Basidiomata of *Chondrogaster pachysporus*. (1. Sulzbacher-192; 2. Sulzbacher-196)

when young, becoming irregularly reticulate at maturity and of variable diameter ( $<3\ \mu\text{m}$ ).

**BASIDIA** monosporic,  $31\text{--}52 \times 4\text{--}16.5\ \mu\text{m}$ , hyaline, subcylindrical, with constricted base and apex, clamp connections common, collapsed in mature specimens. **PERIDIUM** separable from the gleba, 2-layered: a) external layer formed by yellowish brown, thick-walled, clamped hyphae ( $4.2\text{--}11\ \mu\text{m}$  diam.) mixed with abundant soil particles; b) internal layer composed by hyaline, smooth and thin-walled hyphae, compactly interwoven, filamentous to subglobose  $4\text{--}27.5\ \mu\text{m}$  diam. **TRAMA**  $30\text{--}100\ \mu\text{m}$  thick, not gelatinized in young basidiomata, becoming gelatinized in mature specimens, constituted of hyaline, smooth, thin-walled, and compactly interwoven hyphae,  $3.2\text{--}5.5\ \mu\text{m}$  diam., clamp connections rare.

**DISTRIBUTION:** Australia and United States (Bougher & Lebel 2001), North Africa (Lago & Castro 2004), Spain (Lago & Castro 2004, Moreno-Arroyo et al. 2005), Portugal (Calonge & Vidal 2000), France and Italy (Lago & Castro 2004). Probably widespread with eucalypt trees.

**SPECIMEN EXAMINED:** **BRAZIL.** Rio Grande do Sul: Santa Maria, Boca do Monte District, Estação Experimental de Silvicultura-FEPAGRO, 21 April 2009, leg. M.A. Sulzbacher 191 (SMDB 12.920); *ibid.*, 06 May 2009, leg. M.A. Sulzbacher 192 (SMDB 12.921); *ibid.*, 10 July 2009, leg. M.A. Sulzbacher 196 (SMDB 12.922; ICN 154459).

**COMMENTS** — The genus *Chondrogaster* was considered in the past as a member of the *Melanogastraceae* E. Fisch. (Zeller 1949), *Hysterangiaceae* E. Fisch. (Bougher & Lebel 2001), and *Chondrogastraceae* Locq. (Giachini et al. 2000). However, molecular phylogenetic analysis places the genus is currently placed in the *Mesophelliaceae* Jülich of the *Hysterangiales* K. Hosaka & Castellano (Hosaka et al. 2006).

*Chondrogaster pachysporus*, originally described by Maire from Mauritania (Africa), constitutes the type species of the genus, which until recently was considered monotypic (Giachini et al. 2000). Although it is associated with widely cultivated *Eucalyptus* spp. in the world, this species has been poorly documented, probably due to its underground cryptic habit, as most mycologists pay little attention to hypogeous fungi.

As far as we know, this is the first record in South America. *Chondrogaster angustisporus*, which has been reported from southern Brazil, Australia, and Uruguay (Giachini et al. 2000), differs from *C. pachysporus* in the narrower basidiospores ( $10\text{--}15 \times 4\text{--}5\ \mu\text{m}$ ) covered by a less coarse ornamentation and the presence of mostly bisporic basidia within the glebal locules (Lago & Castro 2004). In contrast to our specimens, which were collected under *E. saligna*, *C. angustisporus* has been found under *E. dunnii* Maiden in southern Brazil as well as several other *Eucalyptus* species in Australia (Giachini et al. 2000).

Lupatini et al. (2008) characterized southern Brazilian strains of *C. angustisporus* through mycorrhizal morphotyping and ITS (rRNA) sequences. Their



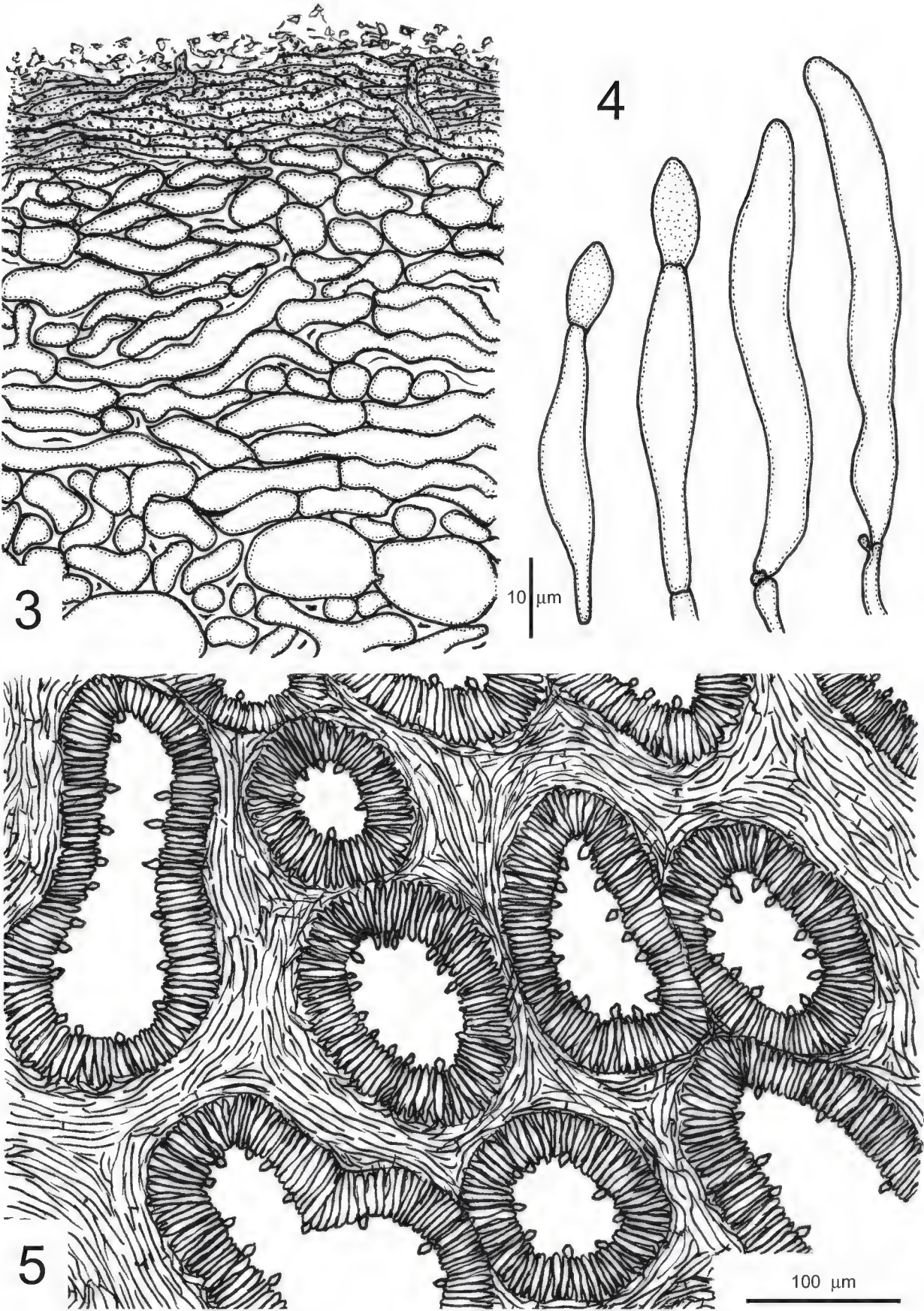


FIG. 3-5. *Chondrogaster pachysporus*. 3. Peridium. 4. Basidia. 5. Gleba structure.

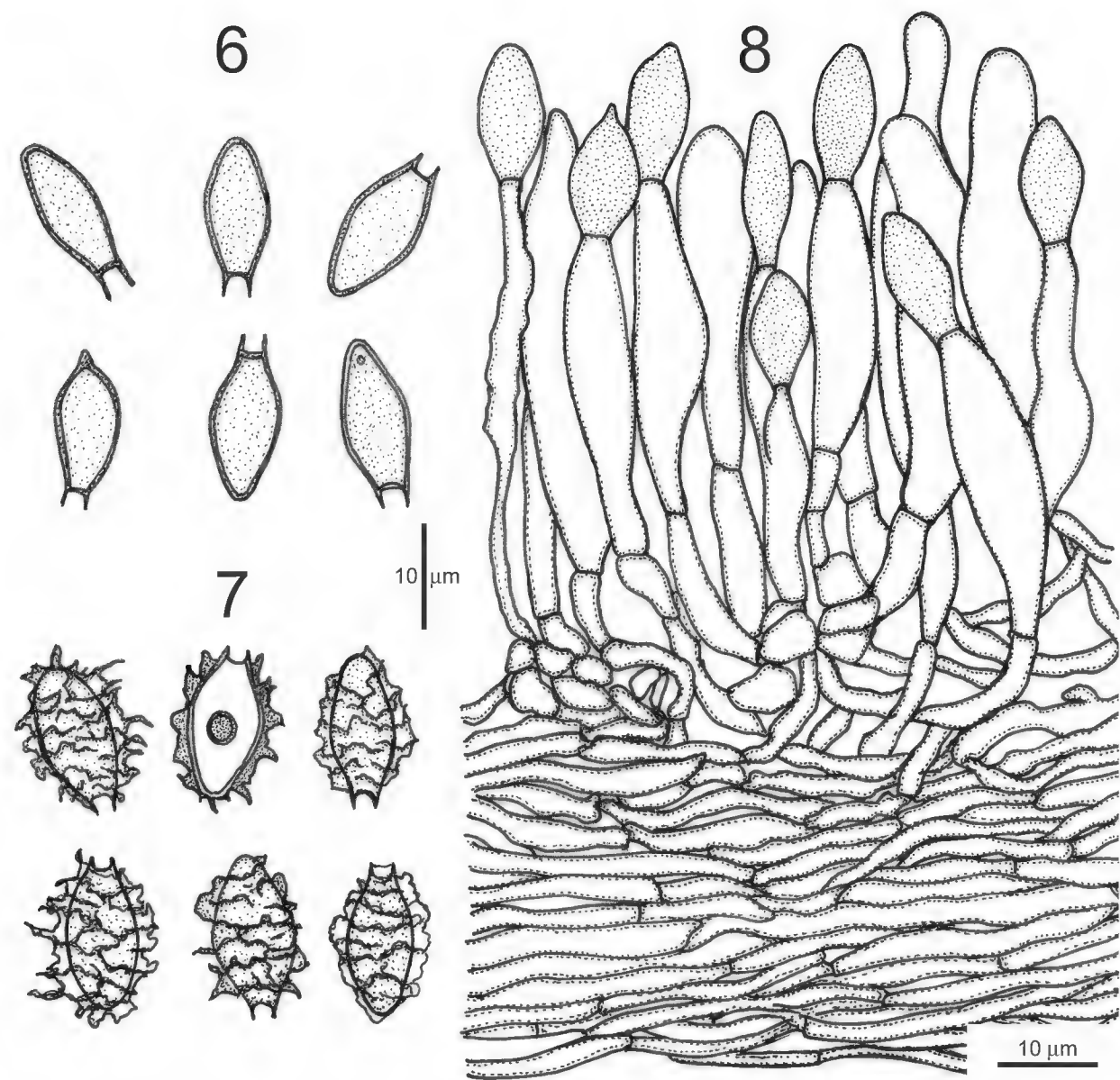


FIG. 6-8. *Chondrogaster pachysporus*.

6. Immature basidiospores. 7. Mature basidiospores. 8 Detail of the hymenium and trama.

results supported strong relationships among other taxa in the gomphoid-phalloid clade (e.g. *Gautieria*, *Gloeocantharellus*, *Gomphus*, *Hysterangium*, *Ramaria*, and *Sphaerobolus*). In two recent molecular phylogeographic studies of *Hysterangiales*, *Andebbia pachythrinx* (Cooke & Masee) Trappe et al. clustered with *C. angustisporus* and *C. pachysporus*, suggesting a close relationship between them (Hosaka et al. 2006, 2008).

The present report from southern Brazil considerably extends the known world distribution of *C. pachysporus*. The new record arises from ongoing investigations of hypogeous fungi associated with *Eucalyptus* in the state of Rio Grande do Sul. As this research progresses, we hope to provide additional data on their diversity and biology.



## Acknowledgments

The authors thank Dr. Eduardo R. Nouhra (Universidad Nacional de Córdoba, Argentina) and Dr. Marisa L. Castro (Universidad de Vigo, Spain) for pre-submission reviews of the manuscript, Dr. Fabrício A. Pedron and MSc. Fábio P. Menezes for their help in soil classification, and CAPES and CNPq (Brazil) for financial support.

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## MYCOTAXON

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***Neobulgaria alba* sp. nov. and  
its *Phialophora*-like anamorph in native forests  
and kiwifruit orchards in New Zealand**

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**Abstract** – Surveys of fungi associated with stained vascular tissue in kiwifruit vines in New Zealand have consistently revealed *Phialophora*-like fungi. Phylogenetic analyses based on DNA sequences have shown most of these to be *Leotiomyces*. The teleomorph of the most common species isolated from stained kiwifruit wood has been found on fallen wood in native forests, and it is described here as *Neobulgaria alba* sp. nov. Other species isolated from kiwifruit wood matched *Cadophora* and *Mollisia* spp. reported from similar symptoms from kiwifruit and other hosts in other countries.

**Key words** – vascular staining, pathogen, *Actinidia deliciosa*, phylogeny, *Phaeoacremonium*

## Introduction

Discoloured and decayed wood is common in the trunks of kiwifruit (*Actinidia deliciosa*) vines more than about 10 years old. The stained wood symptom is similar to that recorded in Italy where a number of fungi including *Phaeoacremonium* spp. and *Phialophora* spp. were isolated from diseased trunks (Di Marco et al. 2000). Surveys in New Zealand of fungi associated with these symptoms revealed several common *Phialophora*-like spp. (e.g. Manning et al. 2003, Manning & Currie 2007). Subsequent DNA sequencing showed them to represent *Leotiomyces*, several matching described or known *Cadophora* spp. (unpubl. data).

Gams (2000) proposed using the genus *Cadophora* to accommodate some of the leotiomycete-related *Phialophora* spp. and Harrington & McNew (2003) clarified the taxonomy of these species. Based on ITS sequences, New Zealand isolates from kiwifruit wood match *Cadophora luteoolivacea* (e.g. Genbank accession HM116748), *Mollisia dextrinospora* (e.g. Genbank accession HM116746), and an apparently unnamed clade that includes “*Cadophora melinii*” sensu Prodi et al. (2008) but is genetically distinct from the ex holotype isolate of *C. melinii* (e.g. Genbank accession HM116752). This last species has been reported from wood and roots of various trees from Europe as Dark Septate Endophyte ITS Haplotype III in Grünig & Sieber (2005; Genbank accession number AY664502) and as *Phialophora malorum* aggregate in Lygis et al. (2005; Genbank accession number AY787725).

*Phialophora*-like anamorphs are spread throughout the *Leotiomyces*. Examples discussed by Gams (1980) included representatives from the families *Helotiaceae*, *Dermateaceae*, *Hyaloscyphaceae*, and *Sclerotiniaceae*. Later authors have linked the *Phialophora*-like genera *Catenulifera* and *Phialocephala* to the leotiomycete genera *Hyphodiscus* (Hosoya 2002, Untereiner et al. 2006) and *Mollisia* (Grünig et al. 2009) respectively.

In this paper we report a *Phialophora*-like leotiomycete anamorph associated with a new *Neobulgaria* sp. The anamorph has been found in cultures grown from ascospores from apothecia collected in native forests and it is also commonly isolated into culture from diseased kiwifruit wood.

## Methods

A survey of kiwifruit (*Actinidia deliciosa*) in 38 orchard blocks in South Auckland/Waikato (15), Bay of Plenty (15), Hawkes Bay (2), Nelson/Golden Bay (4), and Kerikeri (2) was carried out between 2002 and 2007. Samples of wood from symptomatic and non-symptomatic trunks were taken using a 4 mm diam core borer and small pieces of wood from these placed on Difco potato dextrose agar plates (PDA) with streptomycin and penicillin G added. The fungi isolated were grouped on the basis of cultural appearance and micro-morphology. Representative isolates were stored as a working collection in 10% glycerol at  $-80^{\circ}\text{C}$  and later placed in permanent storage in liquid nitrogen in the ICMP culture collection, Landcare Research, Auckland. DNA sequences were generated from these isolates following the methods below.

Apothecia were collected during a survey of wood rotting fungi in native forests (Paulus et al. 2006). While the collections were still fresh, ascospores were shot from living apothecia onto agar plates; germinating ascospores were transferred to new plates, and following adequate growth, cultures were placed in permanent storage in liquid nitrogen in the ICMP culture collection. Collections were dried and deposited in the New Zealand Fungal Herbarium (PDD). Macroscopic appearance was described from field notes and from dried herbarium material, and microscopic features described following rehydration of herbarium material in 3% KOH with Meltzer's reagent added.

TABLE 1. Collections used to generate DNA sequences for FIG. 2 in addition to those from Wang et al. (2006b), with Genbank accession numbers.

SPECIES*	VOUCHER <sup>a</sup>	SSU, ITS, LSU rDNA	NOTES
* <i>Cadophora luteoolivacea</i>	ICMP 18096	HM116765, HM116748, HM116760	ICMP 17109, 18084, 18085, 18097, 18098, 18099 from <i>Vitis vinifera</i> wood and ICMP 18092 from <i>Actinidia deliciosa</i> wood have matching SSU, ITS, and LSU sequences
<i>Hyphodiscus hymeniophilus</i>	MUCL 40275	DQ227258, DQ227258, DQ227258	
<i>Hyphodiscus hymeniophilus</i>	CBS 529.87	—, GU727555, GU727555	
* <i>Mollisia dextrinospora</i>	ICMP 18083	HM116762, HM116746, HM116757	ICMP 17107, 17108, 17110, 17111, 17112 from <i>Actinidia deliciosa</i> wood have matching ITS sequences (other genes not sampled)
* <i>Neobulgaria alba</i>	ICMP 18072	HM116761, HM116745, HM116756	ICMP 17113, 17114, 17115, 18073, 18074, 18075, 18076, 18077, 18078, 18079, 18080, from <i>Actinidia deliciosa</i> wood have matching SSU, ITS, and LSU sequences
* <i>Neobulgaria alba</i>	ICMP 18394, culture from Holotype	HM116781, HM116783, HM116782	ICMP 18395 from fallen wood in native forest has matching SSU, ITS, and LSU sequences
<i>Neobulgaria pura</i>	CUP 063609	DQ257364, DQ257366, DQ257365	
<i>Phaeomollisia piceae</i>	UAMH 10851	EU434866, EU434836, —	

\* Indicates sequences generated as part of this study.  
<sup>a</sup> CUP; Cornell Plant Pathology Herbarium, Cornell University, Ithaca, USA. CBS; Centraalbureau voor Schimmelcultures, Utrecht, The Netherlands. ICMP; International Collection of Microorganisms from Plants, Landcare Research, Auckland, New Zealand. MUCL; Industrial Fungi & Yeasts Collection, Belgian Co-ordinated Collections of Micro-organisms, Belgium. UAMH; University of Alberta Microfungus Collection and Herbarium, Alberta, Canada.

Apothecia were sectioned at about 10 µm thickness using a freezing microtome, and sections were mounted in lactic acid.

DNA was extracted from mycelium from cultures grown from both apothecia and stained wood using REDExtract-N-Amp Plant PCR Kits (Sigma, USA), following manufacturer’s instructions. ITS sequences were obtained following the methods of Johnston & Park (2005). Amplification primers for ITS were ITS1 and ITS4 (White et al. 1990); for small subunit ribosomal DNA were NS1 and NS6 (White et al. 1990), and

for the large subunit ribosomal DNA were LROR and LR5 (Bunyard et al. 1994, Vilgalys & Hester 1990). The sequences newly generated for this paper (TABLE 1) have been deposited in Genbank.

DNA sequences were aligned using Clustal X (Larkin et al. 2007), then checked and edited manually. A Bayesian tree was estimated in MrBayes 3.1.2 (Huelsenbeck & Ronquist 2001, Ronquist & Huelsenbeck 2003) with gaps treated as missing data, using a partitioned model, where each portion of the rDNA was assigned a model selected using the AIC method in MrModelTest 2.3 (Nylander 2004). The model selected for the 18S and 28S rDNA was GTR+I+ $\Gamma$  and for the 5.8S rDNA was SYM+I+ $\Gamma$ . The data set was run with 2 chains for 10 million generations, trees sampled every 1000 generations with a burn-in of 10%. Bayesian posterior probabilities were obtained from 50% majority rule consensus trees. In addition to the species recorded from New Zealand, taxa for the analysis were selected from Wang et al. (2006b) to represent genetic diversity across the *Helotiales*, plus the *Phialophora*-like species cited in Untereiner et al. (2006) and Grünig et al. (2009).

***Neobulgaria alba*** P.R. Johnst., D.C. Park & M.A. Manning, **sp. nov.**

FIG. 1

MYCOBANK MB 518281; GENBANK HM116781, HM116782, HM116783.

*Ab Neobulgaria pura ascoporis* (5–)5.5–6.5  $\times$  3–3.5(–4)  $\mu$ m, *apotheciis brunneis differens*.

TYPELOCALITY: New Zealand, vic. Ruatahuna, Tarapounamu, ridge towards Mangapae on western side of road, on fallen decorticated wood intermixed with *Rosellinia* ascomata, P.R. Johnston (D2022) & B.C. Paulus, 13 Dec 2006, (**Holotype**: PDD 91753; culture grown from holotype, ICMP 18394).

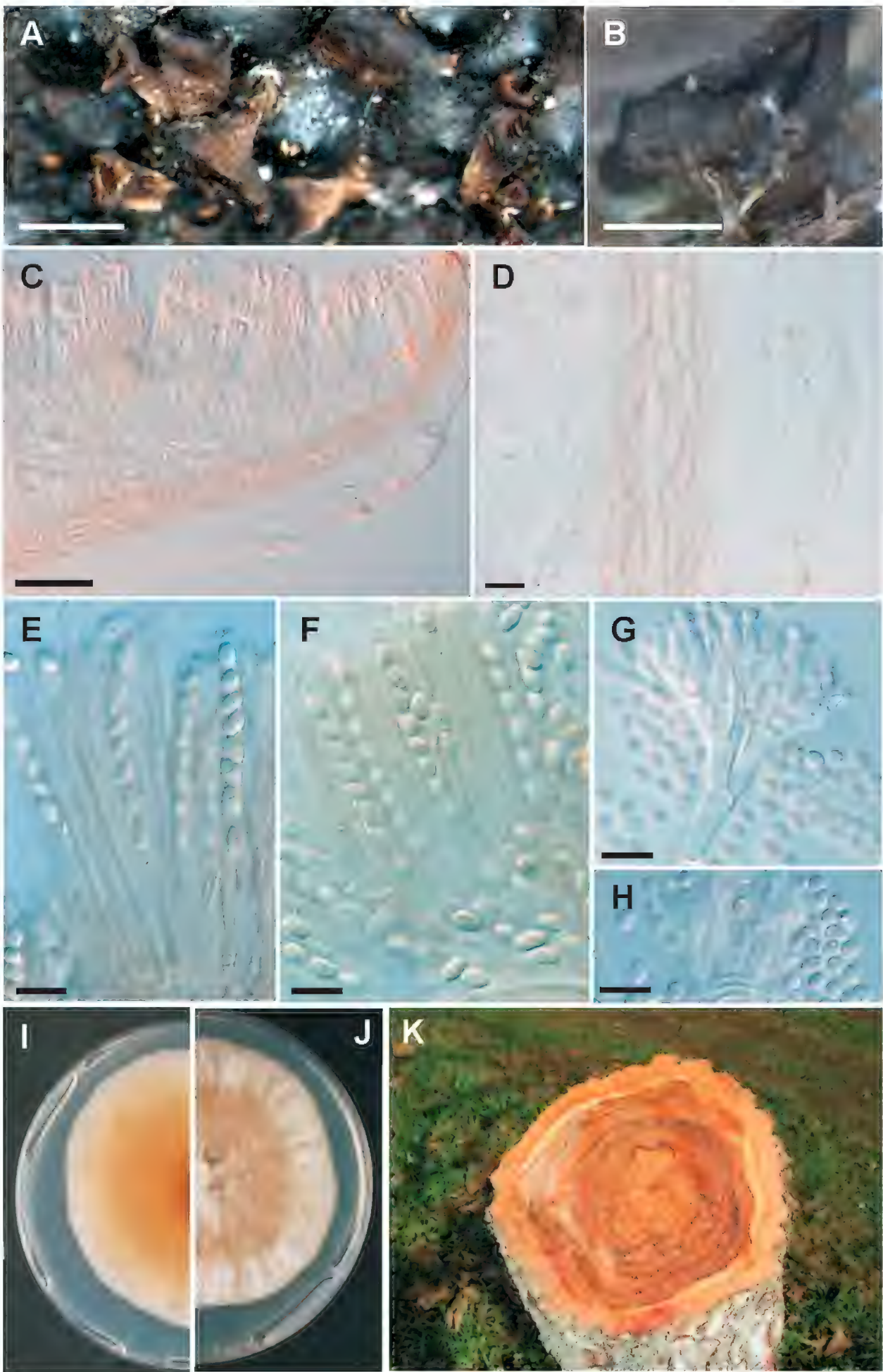
ETYMOLOGY: *alba*, refers to the colour of the colonies on agar isolation plates, paler than most of the other fungi commonly isolated from discoloured vascular tissue.

Apothecia forming on decorticated wood; up to 3 mm diam., substipitate, dull, dark brown, glabrous; when dry receptacle dark brown, hymenium greyish-brown, stipe distinctively flattened. Ectal excipulum with 2 layers; outer layer 35–40  $\mu$ m thick, comprising hyphae 1.5–2  $\mu$ m diam. widely spaced in hyaline gel, oriented more or less parallel to receptacle surface; inner layer 30–35  $\mu$ m thick comprising more or less parallel rows of broad-cylindric cells 5–7  $\mu$ m diam. with walls thin, pale brown, nongelatinous. Medullary excipulum comprising a textura intricata of hyaline hyphae 2  $\mu$ m diam. irregularly oriented and tangled within a hyaline gelatinous matrix. Subhymenium textura

FIG. 1. *Neobulgaria alba* (A, C–F, PDD 91753, holotype; B, PDD 91754; G–J, ICMP 19075). A–B. Apothecia (dry). C. Apothecium in vertical section. D. Apothecium in vertical section, detail of inner and outer ectal excipulum layers with hyphae regularly oriented, and medullary excipulum with hyphae irregularly oriented. E. Paraphyses and asci. F. Asci and ascospores. G. Conidiophores and conidiogenous cells from PDA cultures. H. Conidiogenous cells and conidia from PDA cultures. I. Bottom of 20-day-old colony on PDA. J. Top of 20-day-old colony on PDA. K. Stained kiwifruit wood from which *Neobulgaria alba* was isolated.

Scale bars; A–B = 1 mm; C = 50  $\mu$ m; D–H = 10  $\mu$ m.





intricata, nongelatinous, comprising hyphae with pale brown cell contents. Paraphyses 2–2.5 µm diam., undifferentiated at apex, about same length as asci. Asci 70–110 × 5–6 µm, cylindric, tapering slightly to the subtruncate apex, wall thickened at apex, amyloid plug in the inner half of the wall, more intensely blue to the inside of the wall, 8-spored, uniseriate. Ascospores (5–)5.5–6.5 × 3–3.5(–4) µm, broad-elliptic, symmetrical in both planes, 0-septate, hyaline.

Anamorph in culture. Cultures on Difco PDA 60–70 mm after 20 days, aerial mycelium white, fine, cottony, quite sparse, pale brown agar surface visible through the mycelium. Culture pale yellow-brown in reverse, paler towards the more or less entire margin. Numerous small drops of more or less colourless conidial ooze scattered across surface of colony. Conidiophores penicillate, hyaline, cylindric basal cell 10–15 × 3–5 µm, with 3–4 levels of 2–3 times branching, cylindric cells arising from the basal cell, ending in a terminal conidiogenous cell. Conidiogenous cells 6–8 × 2.5–3.5 µm, more or less cylindric, tapering near apex, with single, apical conidiogenous locus, wall thickened at conidiogenous locus and with flaring collarete. Conidia hyaline, broadly ovate to subglobose, 4.5–5 × 3–3.5 µm.

ADDITIONAL SPECIMENS EXAMINED — New Zealand: Gisborne: vic. Ruatahuna, Te Waiiti, on fallen decorticated wood, P.R. Johnston (D2031) & B.C. Paulus, 4 Dec 2006, (PDD 91754, ICMP 18395). Buller: vic. Reefton, Maimai Creek, on decorticated wood in running water, P.R. Johnston (D1377), 4 Oct 1998 (PDD 70861). Auckland: Waiuku, on *Actinidia deliciosa* 'Hayward', swollen trunks of living vines, M.A. Manning (MM42), 21 Mar 2002 (ICMP 18072); Waiuku, on *Actinidia deliciosa* 'Hayward', swollen trunks of living vines, M.A. Manning (MM43), 19 Mar 2002 (ICMP 18073); Waiuku, on *Actinidia deliciosa* 'Hayward', swollen trunks of living vines, M.A. Manning (MM44), 15 Mar 2002 (ICMP 18074); South Auckland, on *Actinidia deliciosa* 'Hayward', swollen trunks of living vines, M.A. Manning (MM48), 22 Mar 2002 (ICMP 18077); South Auckland, on *Actinidia deliciosa* 'Hayward', swollen trunks of living vines, M.A. Manning (MM49), 22 Mar 2002 (ICMP 18077); Patamahoe, on *Actinidia deliciosa* 'Hayward', swollen trunks of living vines, M.A. Manning (MM421), 22 Mar 2002 (ICMP 18077). Nelson, on *Actinidia deliciosa* 'Hayward', swollen trunks of living vines, M.A. Manning (MM101), 8 Dec 2004 (ICMP 18080). Waikato, Pukekawa, on *Actinidia deliciosa* 'Hayward', swollen trunks of living vines, M.A. Manning (MM46), 19 Mar 2002 (ICMP 18075); Pukekawa, on *Actinidia deliciosa* 'Hayward', swollen trunks of living vines, M.A. Manning (MM47), 19 Mar 2002 (ICMP 18076). Bay of Plenty: Te Puke, on *Actinidia deliciosa* 'Hayward' trunk decay, M.A. Manning (MM468), 13 Jul 2006 (ICMP 17114); Tauranga, on *Actinidia deliciosa* 'Hayward' trunk decay, M.A. Manning (MM493), 23 Mar 2007 (ICMP 17115); Te Puke, on *Actinidia deliciosa* 'Hayward' trunk decay, M.A. Manning (MM494), 20 Mar 2007 (ICMP 17113).

NOTES — Eleven species have been described in *Neobulgaria* (Index Fungorum 2010) — *N. caliciformis* Killerm., *N. faginea* (Peck) Raitv., *N. foliacea* (Bres.) Dennis, *N. lilacina* (Wulfen) Dennis, *N. margaritoidea* Killerm., *N. orientalis* Raitv. & Bogacheva, *N. parvata* V.P. Tewari & Ram N. Singh, *N. premnophila* Roll-Hansen & H. Roll-Hansen, *N. pura* (Pers.) Petr., *N. rupicola* V.P. Tewari

& Ram N. Singh, and *N. undata* (W.G. Sm.) Spooner & Y.J. Yao. The decision to describe *N. alba* as new is based in part on its genetic distinctness from collections putatively representing three north-temperate species, *N. pura* (DQ257364), *N. foliacea* (NPU45443), and *N. premnophila* (NPU45445), and in part on its geographic range. The 18S rDNA sequences of the three Northern Hemisphere collections differ from each other by 1–2 bases, whereas *N. alba* is 8 base pairs different.

Based on published descriptions, all of the described species differ from *N. alba* variously in ascospore size and septation, apothecial shape and size, and pigmentation in culture (Dennis 1956, 1971, Killerman 1929, Lizoň et al. 1998, Raitviir & Bogacheva 2007, Roll-Hansen & Roll-Hansen 1979, Seaver 1961, Tewari & Singh 1975). From the illustrations of Tewari & Singh (1975), *N. rupicola* appears to have the apothecial anatomy of *Ascocoryne*. From the description provided by Dennis (1954), based in part on material collected in tropical America, *N. alba* is very similar to *Ombrophila microspora* (Ellis & Everh.) Sacc. & P. Syd. However, Dennis (1954) described the asci of *O. microspora* as being barely thickened at the apex and as having an indistinct amyloid reaction. Lizoň et al. (1998) regarded *O. microspora* as a synonym of *N. pura*, a good illustration that species limits within the genus remain confused. Genetic studies on authentically identified specimens are needed to resolve the relationships between species of this genus, and of those that have been placed in *Ombrophila*.

## Discussion

Common in living wood of mature kiwifruit vines but found also in native forests, the genetically distinct *Neobulgaria alba* is assumed to be a native New Zealand species that has moved from natural to human habitats. Data from Johnston (2010) show that many putatively native species of fungi have moved into modified habitats and formed associations with exotic host plants. Although represented by only three collections from native forests, this macroscopically insignificant fungus is assumed to be widespread in New Zealand forests. Its occurrence on kiwifruit suggests that it will have a naturally wide host range. Its biology in native forests is likely to be similar to that in kiwifruit orchards — as well as being apparently saprobic on fallen wood it will probably be found also in association with discoloured wood of living trees.

In New Zealand kiwifruit orchards *N. alba* has been found almost exclusively in association with swollen trunks and stained wood, with only one isolate from symptomless wood (Manning et al. 2003). Following inoculation of healthy wood of kiwifruit with *N. alba*, the fungus was subsequently re-isolated 8 months later from stained tissue surrounding the point of inoculation (unpublished data, M.A. Manning). Less commonly isolated from the same symptoms



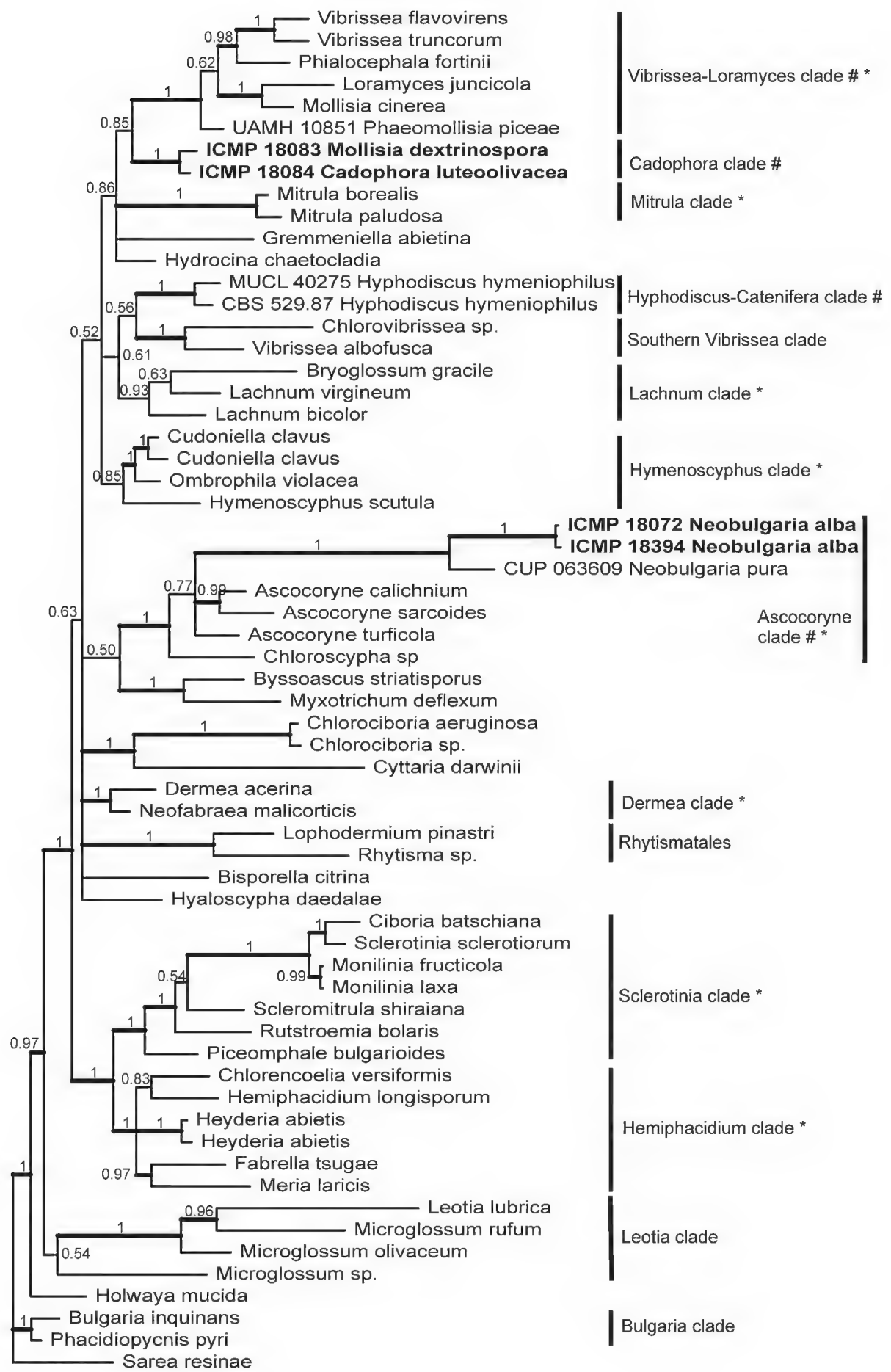
were putatively novel *Phaeoacremonium* spp. (e.g. ICMP 18093, Genbank HM116770, HM116776; ICMP 18094, Genbank HM116771, HM116777) and *Cadophora* spp. (FIG. 2 and TABLE 1). *Cadophora* and *Phaeoacremonium* spp. have been isolated from similarly discoloured wood in Italy, and pathogenicity tests again showed that they caused the symptoms (Di Marco et al. 2008). Field observations suggest that these fungi are unlikely to cause stem death, but they do impact on fruit yield. Fresh weight of fruit from New Zealand vines with disease symptoms averaged 12.6 g less than those from apparently healthy vines (Manning & Currie 2007).

The fungus from New Zealand kiwifruit has in the past been referred informally to “*Phialophora alba*” (e.g. Manning et al. 2003, Prodi et al. 2008). *Phialophora alba* was described from diseased wood by van Beyma (1943). He described a fungus with subglobose conidia,  $3\text{--}4 \times 2.7 \mu\text{m}$ , similar in size to *Neobulgaria alba*, but the ITS sequence from the type specimen (CBS 112.43, Genbank accession number HM116755) showed it to represent a *Paecilomyces* sp.

The genus *Neobulgaria* is characterised by the excipulum having a layer of non-gelatinous, cylindric cells sandwiched between gelatinous tissue to both the inside and outside. The outer gelatinous layer comprises narrow hyphae in a thick gel matrix, oriented more or less parallel to the receptacle surface, whereas the inner layer has hyphae irregular in orientation and less widely spaced in gel. Some species placed in *Ombrophila* have the same excipular structure (e.g. *O. microspora* as illustrated by Dennis 1954) and several authors have discussed the possibility that the two genera may be synonyms (e.g. Carpenter 1981, Dennis 1956, Verkley 1992). Baral & Krieglsteiner (1985) placed the genera in synonymy, recombining the type species of *Neobulgaria* in *Ombrophila*. However, the only genetic data available for the two genera indicate that they should be retained as distinct. *Ombrophila* belongs with some *Hymenoscyphus* spp. in what Wang et al. (2006a, b) suggested could represent a ‘core’ *Helotiaceae* sensu stricto clade, while *Neobulgaria* forms an unnamed clade with other genera with highly gelatinised excipular tissues, including *Ascocoryne* and *Chloroscypha*. Additional *Neobulgaria* sequences generated as part of this study support its position close to *Ascocoryne*.

Phylogenetic relationships of leotiomycete fungi with *Phialophora*-like anamorphs remain poorly resolved (FIG. 2). As suggested by Grünig et al.

FIG. 2. 50% majority-rule consensus phylogenetic tree based on Bayesian analysis of 18S rDNA, 5.8S rDNA, and 28S rDNA regions. Details of taxa with voucher numbers before the names are provided in Table 1, all other taxa are from Wang et al. (2006b). Bayesian posterior probabilities are shown above the edges, and those greater than 95% are indicated with bold lines. Informal clade names marked with \* follow Wang et al. (2006a) and those marked with # are clades containing wood-staining fungi discussed in this paper. *Sarea resinae*, basal to the monophyletic *Leotiomyces* in Wang et al. (2006b) was selected as the outgroup.





(2009), *Phialocephala*, *Acephala*, and their newly described genus *Phaeomollisia*, belong in the *Vibrissea-Loramyces* clade of Wang et al. (2006a). In our analyses, *Cadophora* forms a poorly supported sister relationship with the *Vibrissea-Loramyces* clade, and the position of *Hyphodiscus* remains unresolved amongst a group of the Wang et al. (2006a) clades including the *Vibrissea-Loramyces*, *Mitrula*, *Hymenoscyphus*, and *Lachnum* clades (FIG. 2).

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## MYCOTAXON

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***Chaetospermum setosum* sp. nov.  
from the Western Ghats, India**KUNHIRAMAN C. RAJESHKUMAR, PARAS N. SINGH, LAL S. YADAV,  
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**Abstract** – A new species of *Chaetospermum*, *Ch. setosum*, is described based on the presence of conidiomatal setae and differences in conidial size and shape. This species occurs on *Mangifera indica* (Anacardiaceae) collected from Bhima Shankar forests in the Western Ghats of Maharashtra, India. The presence of conidiomatal setae is a unique character that differentiates this species from related taxa.

**Key words** – anamorphic fungi, *Efibulobasidium*, *Sebacinales*

**Introduction**

During July 2009 a survey was conducted to explore the microfungal diversity in the natural forests of Bhima Shankar situated in the northern part of the Western Ghats, India, at 19°40'00"–19°42'09"N 73°29'16"–73°38'06"E with an altitude of 945 msl. The forest types are mainly evergreen and semi-evergreen with rainfall up to 6000 mm per annum (Janardhanan 1966). An unusual *Chaetospermum* species was found on fallen leaves of *Mangifera indica*. The presence of gelatinous conidiomata, holoblastic sympodial conidiogenesis, and cylindrical, non-septate conidia with tubular appendages are the distinguishing features of the genus *Chaetospermum* (Sutton 1980; Nag Raj 1993). Species of *Chaetospermum* are recorded worldwide as common saprophytes isolated from freshwater and litter. Sequences of two species of *Chaetospermum* suggest that members of this genus are basidiomycetes in the order *Sebacinales* (Rungjindamai et al. 2008). The anamorph-teleomorph relationship between

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*Chaetospermum* and a known species of *Sebacinales*, *Efibulobasidium albescens* (Sacc. & Malbr.) K. Wells, was suggested by Wells & Bandoni (2001) and confirmed recently by Kirschner & Oberwinkler (2009). *Chaetospermum setosum*, which differs from the other five species described in that genus based on the presence of conidiomatal setae, cylindrical or V- and Y-shaped conidia, and number of polar appendages, is described as new to science.

### Materials and methods

Conidiomata of the fungus were isolated from the lower surface of fallen leaves and observed under a Nikon Binocular stereo microscope (Model SMZ – 1500 with Digi-CAM, Japan). The serial dilution method was used to isolate this fungus (Pramer & Schmidt 1965) and the hyphal elements from the growing margin of the pure colonies developing from single spores were transferred to new Potato Dextrose Agar plates (PDA). For morphotaxonomic studies and photomicrographs an Olympus CX-41 (Japan) microscope was used. Conidia, setae, and conidiophores were measured using an ocular micrometer. The growth patterns of the colonies were also studied on different culture media viz. Czapek Yeast Autolysate Agar (CYA), Malt Extract Agar (MEA), Potato Carrot Agar (PCA), and PDA (Himedia Mumbai, India). Development was also observed on modified 2% agar media (2 g crushed autoclaved mango leaves mixed in 2% agar). The specimens were deposited in Ajrekar Mycological Herbarium (AMH) and the culture was accessioned and preserved in National Fungal Culture Collection of India (WDCM-932), Agharkar Research Institute, Pune, India.

### Taxonomic description

***Chaetospermum setosum* Rajeshkumar, S.K. Singh & P.N. Singh, sp. nov.**

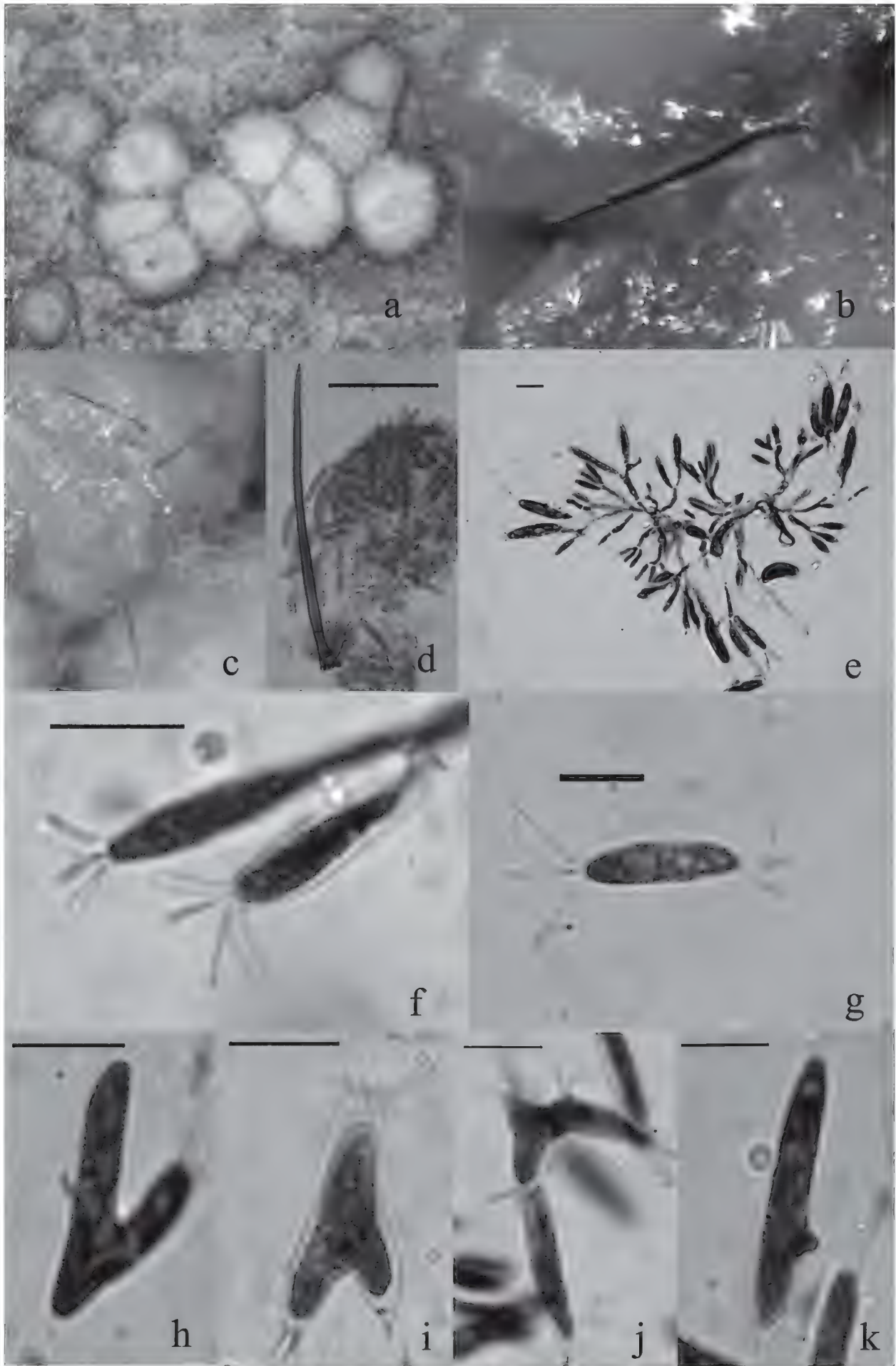
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PLATES 1, 2

*Foliicola. Conidiomata pycnidioidea, 300–750 µm diam., subepidermalia, primum immersa, postremo erumpentia, gelatinosa, nivea ad cremea ubi humida, pallide brunnea ad atrobrunnea ubi sicca. Setae marginales 120–132 µm longae, 5 µm latae ad basim, pallidae ad atrobrunneae, gradatim contractae versus apicem, acuminatae, crassitunicatae, solitariae vel binatim, 1–2-septatae ad basim. Conidiophora ramosa, hyalina, laevia. Cellulae conidiogenae cylindracaе, holoblasticae, sympodiales, conidia maximam partem terminalia interdum lateralialia, 1–4 in fasciculis. Conidia unicellularia, cylindracaа vel variabilia in forma, recta vel curva, hyalina, laevia, guttulata, apice obtusa, 17–29(–40) × 4.4–7 µm. Appendices polares, nonramosae, tubulares, 2–5, praeciupue 3–4, 7.5–12.5 µm longae.*

PLATE 1. *Chaetospermum setosum* (holotype). a. Habit. b–c. Conidiomatal setae. d. Conidiomatal setae and conidia. e. Conidiophores branching and conidiogenous cells. f. Conidial development g. Mature conidia with appendages. h–k. Branched and irregularly shaped conidia with appendages. Bars: d = 50 µm; e–k = 10 µm.





ETYMOLOGY: from Latin *setosus* referring to the conidiomatal setae present in this species.

HOLOTYPE: India, Bhima Shankar, Western Ghats, Maharashtra, on fallen leaves of *Mangifera indica* L. (*Anacardiaceae*) 30 Nov 2009, K.C. Rajeshkumar, AMH 9299. (Ex-type culture NFCCI 1912.)

Foliicolous. Conidiomata pycnidial, 300–750  $\mu\text{m}$  diam., initially immersed, subepidermal, ultimately erumpent, opening by an irregular split in apical wall, gelatinous, pearl white to creamish when moist, pale brown to dark brown when dry, scattered to gregarious, confluent. Setae marginal, 120–132  $\mu\text{m}$  long, 5  $\mu\text{m}$  wide at the base, pale to dark brown, gradually tapering towards the pointed apex, wall thickened, solitary or in pairs, one to two septate at base. Conidiophores arising from innermost wall of conidiomata, branched, hyaline, smooth. Conidiogenous cells cylindrical, holoblastic, sympodial, conidia mostly terminal, sometimes lateral, 1–4 in clusters. Conidia unicellular, cylindrical or variable in shape, sometimes branched, each branch bearing appendages, straight or curved, hyaline, guttulate, smooth-walled, apex obtuse, 17–29 (–40)  $\times$  4.5–7  $\mu\text{m}$  (mean  $24.2 \times 5.3 \mu\text{m}$ ), length-width ratio 4.6:1; appendages polar, unbranched, tubular, 2–5 at each end, usually 3–4, 7.5–12.5  $\mu\text{m}$  long.

TELEOMORPH: Unknown; no sexual state or fungus resembling *Efibulobasidium* was present near the specimen.

Colonies on PDA slowly growing, 15 mm diam. after 7 days and 25 mm diam after 15 days, white, dull white to pale creamish white, with slight ridges and furrows, smooth, flat, margin irregular, aerial mycelium scanty, reverse creamish or dull white. Colonies on MEA slowly growing, 10 mm diam. after 7 days, white, velutinous, smooth, margin irregular, reverse white to off-white. Colonies on PCA fast growing, 60 mm diam. after 7 days, white or off-white, mycelium immersed forming a film over media, flat, margin regular, colonies rounded, reverse white to off-white. Colonies on CYA fast growing, 65 mm diam. after 7 days, creamish white, mycelium immersed forming thin flat colonies, margin regular, reverse white to off-white. Sporulation and conidium morphology on these media were similar to those in nature, but setae were not found.

Sterile seta-like structures developed from the conidiomata in culture grown on modified 2% agar media with crushed autoclaved mango leaves. The sterile hyphae were hyaline or hyaline with dark brown pigmented areas scattered on it, thin-walled, wavy, with a broader base and blunt tip arising from the margins of the gelatinous conidiomata. Sporulation on this medium was poor.

## Discussion

Saccardo (1892) established the genus *Chaetospermum* Sacc. based on *Tubercularia chaetospira* Pat. (Patouillard 1888), now *Ch. chaetosporum* (Pat.)

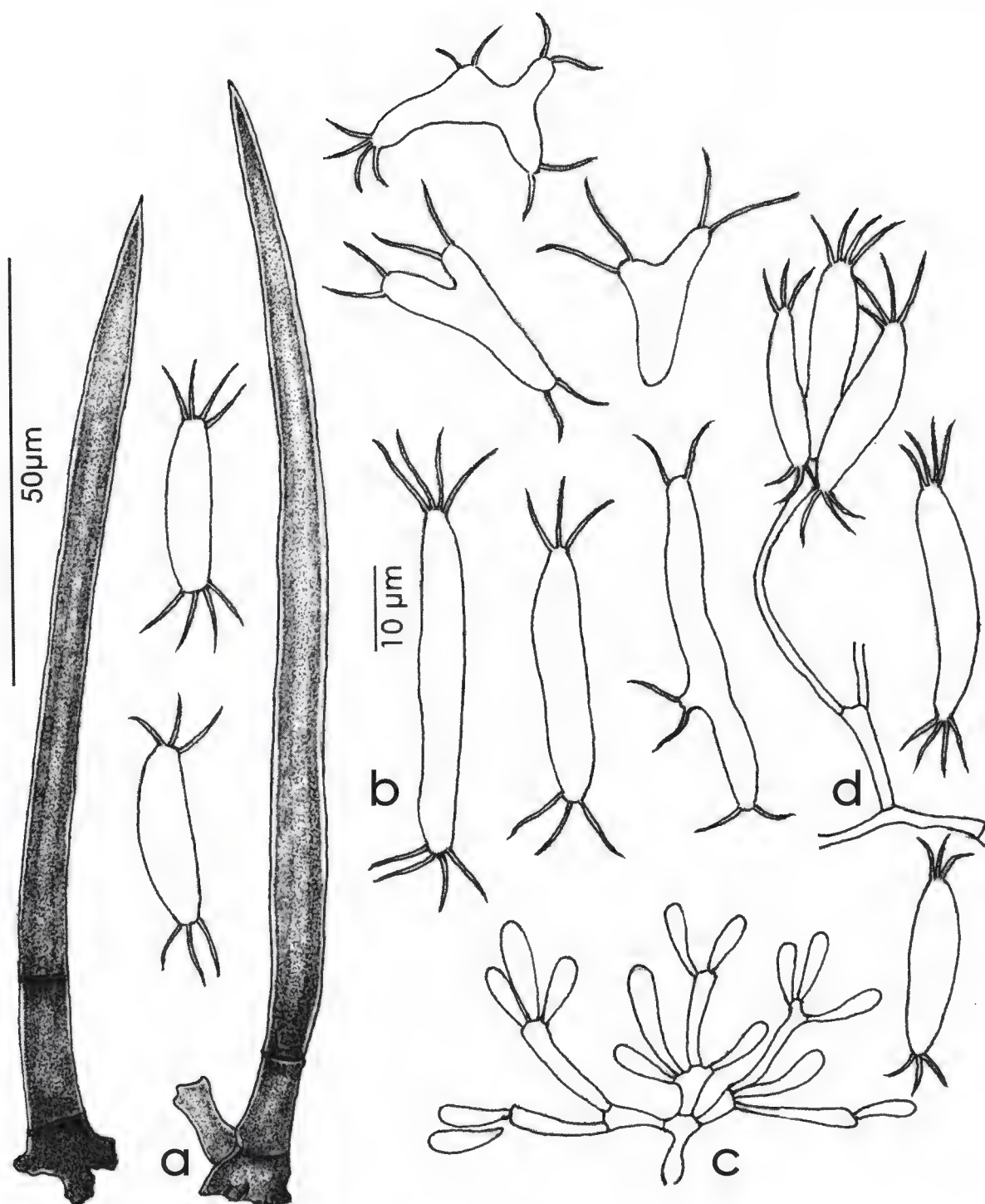


PLATE 2. *Chaetospermum setosum* (holotype).

a. Conidiomatal setae. b. Mature conidia with appendages. c. Conidiophores branching and conidiogenous cells. d. Conidiophores and terminal clusters of conidia.

Bars: a = 50 µm; b–d = 10 µm.

A.L. Sm. & Ramsb. (Smith & Ramsbottom 1914). Saccardo (1892) published a superfluous new name for the type, *Ch. tubercularioides* Sacc., nom. nov., nom. illegit.; this is clearly a homotypic synonym, and not heterotypic as Nag Raj (1993) mistakenly indicated. Nag Raj (1993), who provided the most recent account of the genus *Chaetospermum*, accepted four species: *Ch. chaetosporum*, *Ch. artocarpi* (Nag Raj) Nag Raj, *Ch. camelliae* Agnihothr., and *Ch. gossypinum*



(G.F. Atk.) Nag Raj. He separated these species based on conidial length-width ratio, conidial width, and the position of appendages. He also clarified that conidial appendages in all the taxa in this genus are tubular. Previously, Sutton (1980) had accepted three species of *Chaetospermum* viz., *Ch. carneum* Tassi, *Ch. chaetosporum* and *Ch. gelatinosum* Petch (1917); however, Nag Raj (1993) placed *Ch. gelatinosum* in synonymy with *Mastigonema gelatinosum* (Berk. & Broome) Nag Raj and, following an examination of the type specimen, considered *Ch. carneum* a nomen dubium. Talde (1981) described *Chaetospermum indicum* Talde from India. The type specimen is missing in AMH and not available for re-examination. The description and illustration of *Ch. indicum* suggest that this species is identical with the type species, *Ch. chaetosporum*. Thus, *Ch. indicum* is treated here as a synonym of *Ch. chaetosporum*.

In the present study *Chaetospermum setosum* is proposed as a new species based on its unique morphological characteristics including the presence of conidiomatal setae, variously shaped conidia, and number and origin of the conidial appendages. The presence of conidiomatal setae has not been previously described in *Chaetospermum*. The conidial appendages are polar in *Ch. artocarpi* (as in *Ch. setosum*) but are circumpolar to sub-polar or lateral in the other two species. Although the presence of polar appendages in *Ch. setosum* suggests an affinity with *Ch. artocarpi*, they are more variable, with as many as 5 appendages sometimes present.

*Infundibura adhaerens* Nag Raj & W.B. Kendr. (anamorph of *Helicogloea angustispora* L.S. Olive) is another basidiomycetous anamorph that produces sterile seta-like structures in nature as well as culture. Different authors (Nag Raj & Kendrick 1981; Matsushima 1996; Wu et al. 1997) have given different descriptions for sterile hyphae (setae) in this species. Kirschner (2004), who describes them as hyaline, aseptate, and thick-walled, notes that these differences may be due to intraspecific variation, environmental influences, or aging. *Chaetospermum setosum* also produces setae in nature and sterile hyphae (setae) in culture. In nature the setae are dark brown, erect, with pointed tips, but in culture hyaline or hyaline with dark brown pigmented areas, thin-walled, wavy, and bluntly tipped. This observation indicates that setal characteristics depend on environmental factors and culture conditions.

**Key to species of *Chaetospermum***

- 1. Conidiomata with marginal setae; conidia cylindrical to V- or Y-shaped, appendages polar, 2–5 at each end. . . . . *Ch. setosum*
- 1. Conidiomata without setae; conidia ellipsoidal to cylindrical . . . . . 2
- 2. Appendages polar, 3, rarely 2, appendages on each conidium; conidia 18–26 × 4.5–5.5 µm . . . . . *Ch. artocarpi*
- 2. Appendages circumpolar to subpolar or lateral . . . . . 3

3. Appendages circumpolar to subpolar; conidia  $26\text{--}41 \times 8\text{--}12 \mu\text{m}$   
..... *Ch. chaetosporum*
3. Appendages subpolar or lateral; conidia less than  $8 \mu\text{m}$  wide .....4
4. Appendages  $9\text{--}20 \mu\text{m}$  long; conidial length-width ratio 5.5:1 ..... *Ch. camelliae*
4. Appendages  $18\text{--}20 \mu\text{m}$  long; conidial length-width ratio 6.3:1 ... *Ch. gossypinum*

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## MYCOTAXON

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***Phialophora sessilis*, a species causing  
flyspeck signs on bamboo in China**JIELI ZHUANG<sup>1</sup>, MINGQI ZHU<sup>1</sup>, RONG ZHANG<sup>1</sup>, HUI YIN<sup>1</sup>,  
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**Abstract**—*Phialophora sessilis* is reported and redescribed from China. It is distinguished from the other known species in the genus by reduced, flaring phialidic collarettes and clusters of single-celled conidia. ITS sequence analysis of four strains from Xianning, Hubei, China, attributed to the species shows it to be clearly distinct.

**Key words**—phialide, taxonomy, genetic analysis, sooty blotch, *Gramineae*

**Introduction**

The genus *Phialophora*, which was introduced by Medlar for *P. verrucosa* Medlar isolated from a human skin lesion (Medlar 1915), is currently regarded as a member of *Herpotrichiellaceae* (Haase et al. 1999). It is a little-differentiated genus of more or less pigmented, phialidic hyphomycetes (Hoog et al. 2000). With the addition of numerous species, the genus has become grossly polyphyletic, although some taxa have already been segregated from *Phialophora* into *Cadophora* (*Helotiales*), *Harpophora* (*Magnaporthaceae*), *Lecythophora* (*Coniochaetaceae*) and *Phaeoacremonium* (*Togniniaceae*) (Kirk et al. 2008).

Most *Phialophora* species are common saprobes in soil, wood pulp, and other plant material. Others are more specialized plant pathogens, and human pathogenicity is known for a few species (Gams 2000). *Phialophora sessilis* was first reported by Hoog et al. (1999) from *Picea abies* resin in the Netherlands

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and described in a comparative study of 34 strains belonging to the *Phialophora verrucosa* complex. Additional strains of *Ph. sessilis* originated from forest soils in Sweden, the lichen *Peltigera polydactyla* (Hoog et al. 1999), and marble powder in Italy (Caretta et al. 2006). Important phenetic characteristics of *Ph. sessilis* are dark, slow growing colonies, conspicuous collarettes that are darker than the rest of the phialide and inserted laterally on undifferentiated hyphae, and conidia sometimes inflating and then frequently bearing phialidic collarettes (Caretta et al. 2006).

During a recent survey of host plants for flyspeck fungi in China, two bamboo species were found to be hosts of flyspeck related with *Phialophora sessilis*.

## Materials and methods

### Fungal strains

Four strains were isolated from the culms of two different hosts; isolates ZJ81-D5 and ZJ81-D7 were from *Phyllostachys meyeri*, and ZJ88-B3 and ZJ88-B8 were from *Yushania falcataurita*. Representative dried culture and plant specimens were deposited in the Fungal Herbarium of Northwest A&F University (HMUABO), Yangling, Shaanxi Province, China.

### Isolates

Individual sclerotium-like bodies (Batzler et al. 2005), growing in clusters on bamboo culms, were transferred to slants containing potato dextrose agar (200 g peeled potato, 20 g dextrose, 10 g agar in 1 L water; PDA) and cultured at  $22 \pm 1^\circ\text{C}$  in the dark (Sun et al. 2003). Axenic cultures from slants were transferred to new PDA plates, a sterile cover slip was partially inserted into the agar adjacent to the colony and angled away from the colony at approximately 60 degrees to the agar surface in order to enable the fungus to grow onto the cover slip. Measurements of fungal structures were conducted based on isolates growing on cover slips. Colony descriptions were made after 1 month of growth on PDA plates at  $22 \pm 1^\circ\text{C}$  in darkness.

### DNA extraction, PCR, and sequencing

The protocol of Barnes et al. (2001) was followed to extract genomic DNA from fungal mycelium growing on PDA slants. The primers ITS1-F (Gardes & Bruns 1993) and ITS4 (White et al. 1990) were used to amplify part of the nuclear ribosomal RNA (nrRNA) operon spanning the 3' end of the 18S rRNA gene, the first internal transcribed spacer (ITS1), the 5.8S rRNA gene, the second ITS region (ITS2) and the 5' end of the 28S rRNA gene. The PCR reaction mixture, consisting of 1 unit Taq polymerase, 1× PCR buffer, 2 mM  $\text{MgCl}_2$ , 0.2 mM of each dNTP, 0.4  $\mu\text{M}$  of each primer, and 2  $\mu\text{L}$  template DNA, was made up to a total volume of 25  $\mu\text{L}$  with sterile water. Reactions were performed on a Bio-Rad PCR System PTC-200TM and the cycling conditions were an initial denaturation at  $94^\circ\text{C}$  for 3 minutes followed by 35 cycles of denaturation at  $94^\circ\text{C}$  for 30 seconds, annealing at  $52^\circ\text{C}$  for 30 seconds, extension at  $72^\circ\text{C}$  for 30 seconds, and a final 10-minute extension step at  $72^\circ\text{C}$ . Purifying and automated sequencing with the

primer ITS4/ ITS1-F of the PCR product was performed at Organism Technology Co., Shanghai, China.

Sequence alignment and phylogenetic analyses

The ITS nucleotide sequences generated in this study were added to sequences of six species of *Phialophora* obtained from GenBank ([www.ncbi.nlm.nih.gov](http://www.ncbi.nlm.nih.gov)) (TABLE 1). After importing into BioEdit 5.0.9.1 (Hall 1999), all sequences were pruned to include the complete sequences of ITS1, the 5.8S rDNA gene, and ITS2 to aid alignment. Preliminary alignments of the ITS sequences were conducted using CLUSTAL-X (Thompson et al. 1997), with manual adjustment using BioEdit for visual improvement where necessary.

TABLE 1. Sequences used in the phylogenetic analysis

SPECIES	GENBANK	REFERENCE
<i>Cadophora gregata</i>	AY249070	Harrington & McNew 2003
	AY249071	Harrington & McNew 2003
	U66727	Chen et al. 1996
	U66728	Chen et al. 1996
	U66729	Chen et al. 1996
<i>Cadophora malorum</i>	DQ317328	Arenz et al. 2006
	AF083201	McKemy et al. 2005
	AF083202	McKemy et al. 2005
<i>Phialophora americana</i>	EU514694	Untereiner et al. 2008
	EU514695	Untereiner et al. 2008
	AF050259	Untereiner & Naveau 1999
	AF050260	Untereiner & Naveau 1999
<i>Phialophora europaea</i>	FJ489612	Li et al. 2008
	EF551553	Zeng & Hoog 2008
	EU514698	Untereiner et al. 2008
<i>Phialophora sessilis</i>	AY857542	Prenafeta-Boldu et al. 2006
	AY857541	Prenafeta-Boldu et al. 2006
	DQ363414	Feldmann et al. 2006
	FJ438386	Diaz et al. 2010
<i>Phialophora verrucosa</i>	DQ404353	Prodi et al. 2008
	EU514701	Untereiner et al. 2008
	AF050282	Untereiner & Naveau 1999
	AF050281	Untereiner & Naveau 1999
<i>Pyrenopeziza revincta</i>	AJ430224	Vralstad et al. 2002
ZJ81-D5	GU981734	This paper
ZJ81-D7	GU981735	This paper
ZJ88-B3	GU981736	This paper
ZJ88-B8	GU981737	This paper

Maximum parsimony (MP) analysis was carried out using PAUP 4.0b10 (Swofford 2001). Heuristic searches were conducted with a 1000 random taxa addition and tree bisection-reconnection (TBR) as the branch-swapping algorithm. All characters were unordered and of equal weight and gaps were treated as missing data. Branches of zero length were collapsed and all multiple, equally parsimonious trees were saved. Measures calculated for parsimony included tree length, consistency index, retention index, and rescaled consistency index (TL, CI, RI and RC, respectively). To assess the robustness of clades and internal branches, a strict consensus of the most parsimonious trees was generated and a bootstrap analysis of 1000 replications was performed. The outgroup was *Pyrenopeziza revincta*.

## Results

### DNA phylogeny

Four isolates (ZJ81-D5, ZJ81-D7, ZJ88-B3, ZJ88-B8) were obtained from bamboo. The sequences were deposited in GenBank (ZJ81-D5 = GU981734, ZJ81-D7 = GU981735, ZJ88-B3 = GU981736, ZJ88-B8 = GU981737). Sequences for the ribosomal DNA ITS region (ITS1, 5.8S rDNA gene, ITS2) for each isolate, and related sequence data from GenBank, were used to construct a strict consensus tree with tree length (TL) = 431, consistency index (CI) = 0.8770, retention index (RI) = 0.9671, and rescaled consistency index (RC) = 0.8482 (FIG. 1). Two major clades were resolved in the MP trees. One clade, with 100% bootstrap support, contained two species, *Cadophora gregata* and *C. malorum*. The other major clade (100% bootstrap) consisted of four subclades containing isolates of *P. sessilis*, *P. europaea*, *P. americana*, and *P. verrucosa*. Our four isolates — ZJ81-D5, ZJ81-D7, ZJ88-B3, ZJ88-B8 — clustered together with *P. sessilis* with a 100% bootstrap value, indicating that they probably represent the same species.

### Taxonomy

DESCRIPTION (FIG. 2): HYPHAE initially somewhat torulose, later regularly tubular. Expanding hyphae 1.2–2.7  $\mu\text{m}$  wide, smooth-walled; hyphal cells sometimes inflated to 3.8–7.5  $\mu\text{m}$  wide. PHIALIDES mostly intercalary. COLLARETTES distinct from the rest of the phialide, mostly sessile on undifferentiated hyphae, scattered and independent from placement of septa, triangular to funnel-shaped, up to 1.5  $\mu\text{m}$  long and about 1.5  $\mu\text{m}$  wide at the often somewhat flaring opening. CONIDIA subhyaline, smooth-walled, obovoidal to ellipsoidal, 3.0–7.3  $\times$  2.0–4.5  $\mu\text{m}$ . Conidia in slimy heads, 5.3–8.2  $\times$  6.5–8.4  $\mu\text{m}$ . CHLAMYDOSPORES absent.

SPECIMENS EXAMINED: On *Phyllostachys meyeri* McClure (Gramineae): China, Hubei, Xianning, Qianshan National Forest Park, 29°48'N 114°96'E, alt. 46 m, 16 Oct. 2008, J.L. Zhuang & H.L. Yang, HMUABO 20581 (with dried culture), culture ZJ81-D5 and ZJ81-



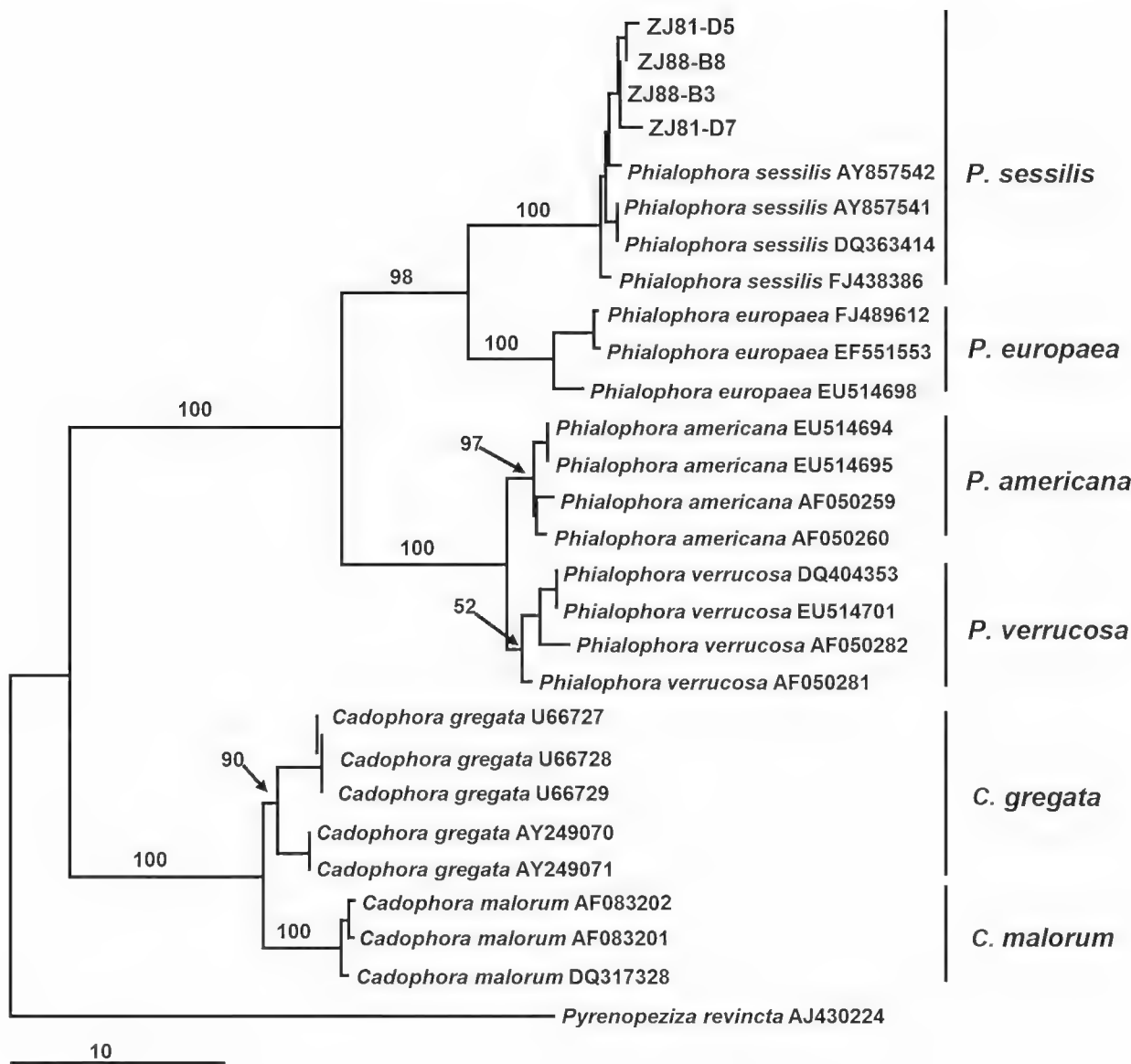


FIG. 1 The parsimony tree (TL = 431, CI = 0.8770, RI = 0.9671, RC = 0.8482) derived from a heuristic search option in PAUP version 4.0b10 with 1000 randomizations of sequence input orders and 1000 bootstrap replications using the data set of ITS1, 5.8S and ITS2. Bootstrap values higher than 50% are indicated above or below the tree branches. The tree was rooted to *Pyrenopeziza revincta*.

D7; on *Yushania falcataaurita* Hsueh & T.P. Yi (*Gramineae*): China, Hubei, Xianning, Qianshan National Forest Park, 29°48'N 114°96'E, alt. 53 m, 16 Oct. 2008, J.L. Zhuang & H.L. Yang, HMUABO 20588 (with dried culture), culture ZJ88-B3 and ZJ88-B8.

CULTURAL CHARACTERISTICS: Colony diameter after 1 month on PDA at 22 ± 1°C reached 20 mm with even margins and rough, farinose aerial hyphae; colony centers were purplish gray and outer zones olivaceous black. On OA similar, colony reaching 23 mm diameter, flat, spreading, with sparse aerial mycelium, surface olivaceous black, but colony color lighter.

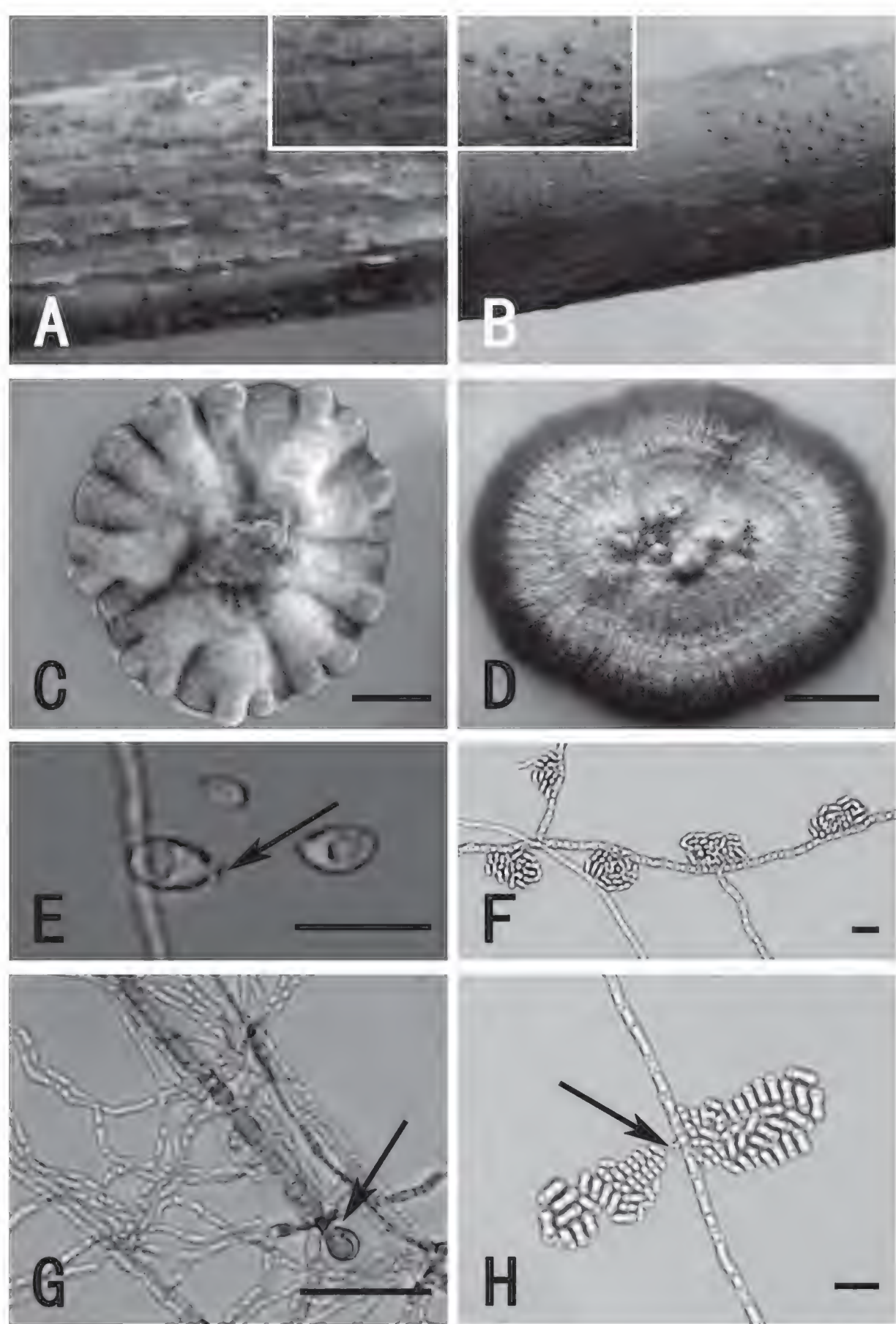


FIG. 2 *Phialophora sessilis* (ZJ81-D5). A. Signs on *Yushania falcataaurita* with close-up view. B. Signs on *Phyllostachys meyeri* with close-up view. C. Colony on PDA after 30 days. D. Colony on OA after 30 days. E. Conidia with open collarette (arrow). F. Conidia and hyphae. G. Inflated hyphal cells (arrow). H. Phialide (arrow) bearing conidia. Bars (C–D) = 0.5 cm. Bars (E–H) = 10  $\mu$ m.

**HOST CHARACTERISTICS:** On *Yushania falcataurita*, no visible mycelial mat with shiny, black, flattened sclerotium-like bodies, round to irregular (130–470 µm diam), scattered distribution over the entire surface of the culm, densely arranged 3–8/mm<sup>2</sup> (FIG. 2A). On *Phyllostachys meyeri* similar, but sclerotium-like bodies were larger (430–680 µm diam), sparse, gathered in clusters on the culm, densely arranged 0.5–1.5/mm<sup>2</sup> (FIG. 2B). The flyspeck on bamboo did not damage the plants, but greatly reduced their ornamental and retail value. As a result, these fungi can cause significant economic losses to producers of these plants.

## Discussion

Our four isolates are morphologically similar to *Phialophora sessilis* de Hoog, and despite minor differences, identity with this species was well supported by the ITS data. The same fungus caused somewhat different signs on each hosts. This phenomenon occurs in other so-called flyspeck species as well, for example, *Dissoconium mali* produced colonies with flyspeck morphology on persimmon fruit (Sun et al. 2008), but colonies with sooty blotch morphology (dark mycelial matrix) on apple fruit (Zhang 2007). It is possible that host-based morphological plasticity may also occur in other fungi in the sooty blotch and flyspeck complex.

Based on the ITS sequence analysis and morphological comparison, we identified the four isolates as *Phialophora sessilis*, which represents a new record for China. This study is also the first report of *P. sessilis* from bamboo.

## Acknowledgments

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## MYCOTAXON

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**Two new anamorphic fungi from Cuba:  
*Endophragmiella profusa* sp. nov. and  
*Repetoblastiella olivacea* gen. & sp. nov.**

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**Abstract** — *Endophragmiella profusa* sp. nov., on submerged decaying leaves of *Bucida palustris* and on bark of decaying nuts of *Couroupita guianensis*, and *Repetoblastiella olivacea* anam. gen. & sp. nov., on bark of decaying nuts of *Couroupita guianensis* from tropical forests in Cuba, are described and illustrated. *Endophragmiella profusa* is distinguished by obovoid, clavate, pyriform to slightly turbinate, 3–5-septate, mostly 5-septate, dark brown, and smooth conidia. *Repetoblastiella olivacea* is characterized by inconspicuous conidiophores and monoblastic, determinate conidiogenous cells that bear cylindrical, multi-septate, olivaceous to pale olivaceous brown conidia—repeatedly blastocatenate and forming several irregular chains from several indeterminate cells across the length of the conidial body.

**Key words** — freshwater fungi, hyphomycetes, systematics

## Introduction

Over thirty anamorphic fungi were collected in Cuba during two mycological surveys of microfungi from tropical plant material (mainly *Bucida palustris*

leaf litter and bark of decaying nuts of *Couroupita guianensis*), in several undisturbed forests of Camagüey and Ciudad de La Habana provinces. Among them, two conspicuous fungi appeared to be new to science and therefore they are described and illustrated here.

### Materials and methods

Samples of submerged plant material were collected during expeditions in 1999 to “Los Cangilones” pool along the Maximo River (Camagüey), and in 2001 to a forest in Santiago de Las Vegas (Ciudad de La Habana). Individual collections were placed in paper bags and taken to the laboratory as described by Castañeda (2005). They were incubated in Petri dishes at 25°C placed in a moist chamber composed of plastic containers (50 L capacity) with 200 ml of sterile water plus 2 ml of glycerol, and examined at regular intervals for the presence of microfungi. Mounts were prepared using polyvinyl alcohol-glycerol (8.0 g in 100 ml of water, plus 5 ml of glycerol). Measurements were made at a magnification of  $\times 1000$ . Micrographs were obtained with a Zeiss Axioskop 40 microscope.

### Taxonomy

*Endophragmiella profusa* R.F. Castañeda, M. Stadler & Gené, sp. nov.

FIG. 1

MYCOBANK MB 518344

COLONIAE in substrato naturali effusae, pilosae, profusae, atrobrunneae, amphigenae. CONIDIOPHORA conspicua, mononemata, simplicia, erecta, recta, cylindrica, 3–7-septata, levia, brunnea, apice versus pallidiora vel subhyalina, 45–170 longa, plus minusve radiatim lobata ad basim, 5–10  $\mu\text{m}$  lata, plerumque simplicia, interdum ad apicem ramosa cum ramis sessilibus, conidiophoris secundariis mutata. CELLULAE CONIDIOGENAE hologenosae, loco fertili uno quaeque indutae, terminales, indeterminatae, cum 2–8 proliferationibus percurrentibus, 9–24  $\times$  2.0–3.5  $\mu\text{m}$ , pallide brunneae vel subhyalinae; in conidiophoris incorporatae. Secedentia conidiorum rhexolytica. CONIDIA solitaria, obovoidea, clavata, pyriformia usque ad turbinata, rotundata ad apicem, conico-truncata ad basim, (2–)3–5-septata, plerumque 5-septata, 22–35  $\times$  7–9  $\mu\text{m}$ ; sicca, laevia, atrobrunnea, sed cum cellulis basalibus dilute brunneis vel subhyalinis, 5–10  $\times$  4–7  $\mu\text{m}$ ; ad basim reliquiis ab partem superiorem cellulae conidiogenae, fimbriata, 1.5–4.0(–8.0)  $\mu\text{m}$  longis praedita. Teleomorphosis ignota.

TYPE: CUBA. CAMAGÜEY: LOS CANGILONES POOL ALONG THE MAXIMO RIVER, 21°35'N; 77°42'W, on submerged decaying leaves of *Bucida palustris* Borhidi & O. Muñiz (*Combretaceae*), 7.II.1999. R.F. Castañeda & J. Cano (Holotype: MUCL 41853).

ETYMOLOGY: Latin, *profusa*, meaning extended, spread out, and referring to the colony.

COLONIES on the natural substratum effuse, hairy, profuse, amphigenous, dark-brown. MYCELIUM superficial and immersed, composed of septate, branched, smooth-walled, brown hyphae, 1–2  $\mu\text{m}$  diam. CONIDIOPHORES macronematous, mononematous, simple or rarely with a branch near the apex, erect, straight,

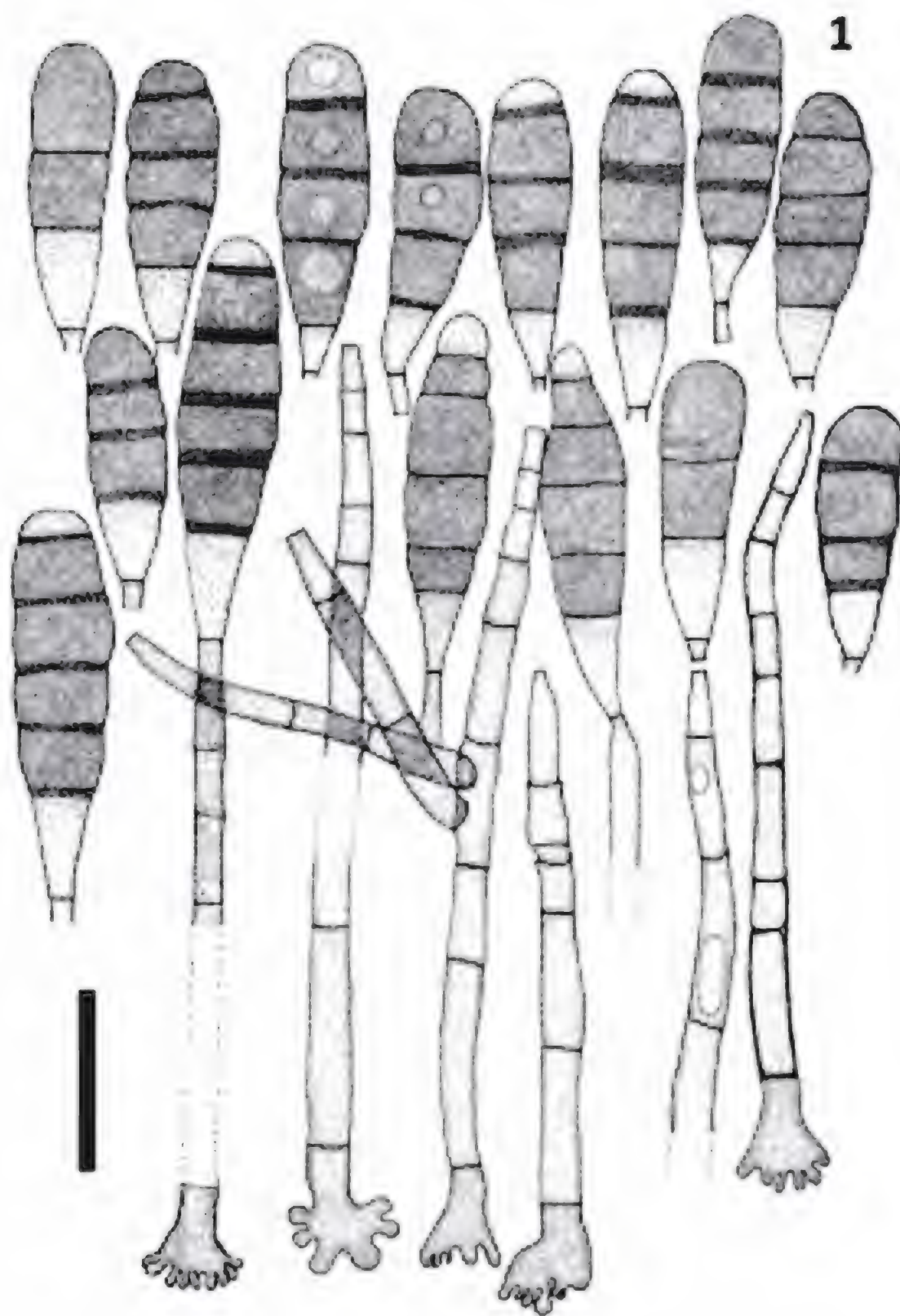


FIG. 1. *Endophragmiella profusa*, drawings from holotype (MUCL 41853).  
Conidiophores, conidiogenous cells, and conidia. Scale bar = 10  $\mu$ m.

cylindrical, 3–7-septate, smooth, 45–170  $\mu$ m tall, basal cell radially lobed, 5–10  $\mu$ m wide, brown at the base, pale brown towards the apex. CONIDIOGENOUS CELLS monoblastic, integrated, terminal, cylindrical, indeterminate, pale brown, with 2–8 enteroblastic percurrent proliferations, 9–24  $\times$  2.0–3.5  $\mu$ m.



Conidial secession rhexolytic. CONIDIA solitary, acrogenous, obovoid, clavate, pyriform to turbinate, rounded at the apex, conical-truncate at the base, (2–)3–5-septate, mostly 5-septate,  $22\text{--}35 \times 7\text{--}9 \mu\text{m}$ , dry, smooth-walled, dark brown, with the basal cell pale brown or subhyaline,  $5\text{--}10 \times 4\text{--}7 \mu\text{m}$ , fimbriate, with a conspicuous basal fringe,  $1.5\text{--}4.0\text{--}(8.0) \mu\text{m}$  long. Teleomorph unknown.

ADDITIONAL SPECIMENS EXAMINED: CUBA. CIUDAD DE LA HABANA: SANTIAGO DE LAS VEGAS,  $22^{\circ}58' \text{N}$ ;  $82^{\circ}20' \text{W}$ , on bark of decaying nuts of *Couroupita guianensis* Aubl. (*Lecythydaceae*), 2.VII.2001. R.F. Castañeda, INIFAT C01/54-4.

NOTES. The genus *Endophragmiella* was erected by Sutton (1973) for *E. pallescens* B. Sutton, the type species, and is distinguished by macronematous, mononematous conidiophores usually unbranched or rarely with a few branches and conidiogenous cells with repeatedly conspicuous enteroblastic percurrent proliferations. Conidial secession is rhexolytic and the conidia mostly bear a very pale pigmented portion of the conidiogenous cell or basal frill (Hughes 1979, Kirk 1985, Wu & Zhuang 2005). *Endophragmiella profusa* slightly resembles *E. tenuis* R.F. Castañeda (Castañeda 1987), but the latter has clavate, obtuse to rounded apex,  $37\text{--}50 \times 5\text{--}6.5 \mu\text{m}$ , brown, (3–6)-septate, mostly 4-septate conidia with pale brown ends.

***Repetoblastiella*** R.F. Castañeda, Minter & M. Stadler, *anam. gen. nov.*

MYCOBANK MB 518342

*Fungus anamorphicus. COLONIAE in substrato naturali pilosae, caespitosae vel funiculosae usque ad arachnoides, effusae, atrovirides, olivaceae vel brunneae. Mycelium partim superficiale et partim in substrato immersum. CONIDIOPHORA micronematosa, mononematosa, simplicia vel ramosa, septata, brunnea vel olivacea, levia vel verrucosa, interdum ad cellulam conidiogenam reducta. CELLULAE CONIDIOGENAE monoblasticae, terminales, determinatae, nonnunquam polyblasticae sympodiales. CONIDIORUM SECESSIO schizolytica. CONIDIA blasto-catenulata, cylindrica, oblonga usque ad longa bacilliformia, multiseptata, olivacea vel brunnea, laevia vel verrucosa; conidia quaeque quaque in cellula facilitatem induta nova producere conidia in catenis.*

ETYMOLOGY: Latin, *repetite*, meaning repeatedly; *blastiella* referring to the blastic mode of conidium ontogeny.

SPECIES TYPICA: *Repetoblastiella olivacea* R.F. Castañeda, Minter & M. Stadler

Anamorphic fungi. COLONIES on the natural substratum hairy, caespitose, funiculose to arachnoid, effuse, dark green, olivaceous or brown. Mycelium superficial and immersed. CONIDIOPHORES micronematous, mononematous, simple or branched, septate, brown or olivaceous, smooth or verrucose, sometimes reduced to a single conidiogenous cell. CONIDIOGENOUS CELLS monoblastic, terminal, determinate; sometimes polyblastic with sympodial proliferations. CONIDIAL SECESSION schizolytic. CONIDIA cylindrical, oblong to bacilliform, multi-septate, olivaceous to pale olivaceous-brown or brown, repeatedly and randomly blastocatenate, forming several irregular chains from



several indeterminate cells across the length of the conidial body. Teleomorph unknown.

NOTES. Several previously described anamorphic genera commonly found in aquatic habitats form somewhat branched or blastocatenulate conidia which originate in a predictable fashion from identifiable cells of the conidial body, and the conidial chains are more or less predictable and stable for each species. Most of these fungi, such as *Dendrospora* Ingold and *Varicosporium* W. Kegel,



FIGS 2–3. *Repetoblastiella olivacea*, photographs from holotype (INIFAT C00/36–3). Conidiogenous cell and conidia forming repeatedly and randomly chains. Scale bars = 10  $\mu$ m.

lack pigmentation. *Catenulostroma* Crous & U. Braun, *Lylea* Morgan-Jones, *Trimmatostroma* Corda, and *Xylomyces* Goos et al. have also micronematous or undifferentiated conidiophores, and conidia are formed in branched chains, sometimes from several cells across the body of each “ramoconidium” similarly to *Repetoblastiella*. In *Catenulostroma*, however, conidiogenous cells are holoblastic-thalloblastic, meristematic and conidial chains are basipetal, *Lylea* has distoseptate conidia forming chains from apical and subapical cells of each ramoconidium, and *Trimmatostroma* has thalloblastic conidial ontogeny with evident disarticulation during conidial secession and often dictyoseptate conidia. The conidial chains in *Xylomyces* show restricted growth in relation to the anastomosing process of the assimilative hyphae, but secondary and tertiary conidia originate only from one cell of the parent conidium. The conidial development in the present genus is enigmatic in the remarkable ability of each conidium cell to produce another conidium, resulting in colonies that are visually complex and net-like in appearance.



FIG. 4. *Repetoblastiella olivacea*, photograph from holotype (INIFAT C00/36–3). Blastocatenulate conidia. Scale bar = 10 µm.

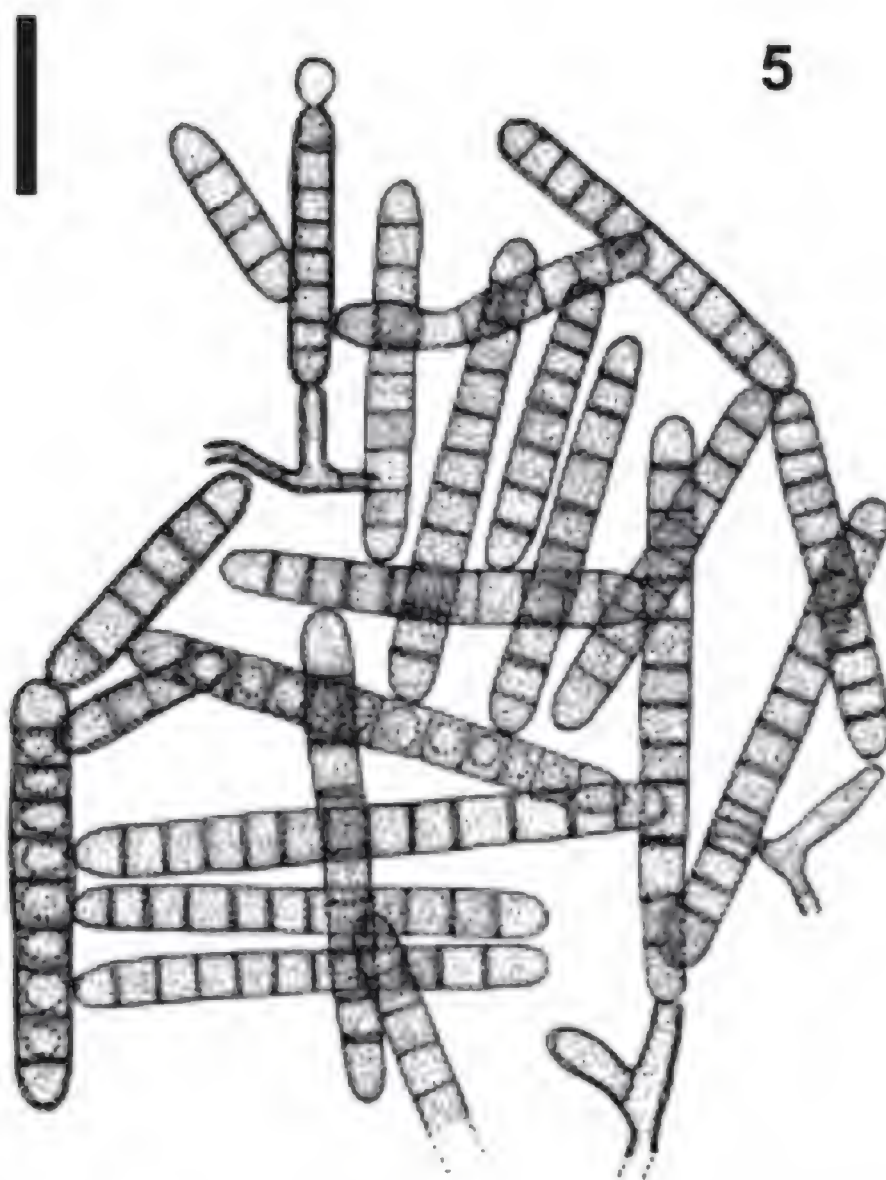


FIG. 5. *Repetoblastiella olivacea*, drawings from holotype (INIFAT C00/36–3). Conidiogenous cells and blastocatenulate conidia. Scale bar = 10  $\mu$ m.

***Repetoblastiella olivacea* R.F. Castañeda, Minter & M. Stadler, anam. sp. nov.**

MYCOBANK MB 518343

FIGS. 2–5

*COLONIAE* in substrato naturali pilosae, funiculosae, effusae, olivaceae. Mycelium partim superficiale et partim in substrato immersum. Hyphae septatae, ramosae, leves, dilute brunneae, 1–2  $\mu$ m diam. *CONIDIOPHORA* micronematosa, mononematosa, simplicia, septata, brunnea vel olivacea, levia plerumque ad cellulam conidiogenam reducta. *CELLULAE CONIDIOGENAE* monoblasticae, terminales, determinatae, 5–10  $\times$  3–6  $\mu$ m, nonnunquam polyblasticae sympodiales. *CONIDIORUM SECESSIO* schizolytica. *CONIDIA* blastocatenulata, cylindrica, oblonga usque ad longa-bacilliformia (2–)8–9(–16)-septata, levia, olivacea, atroviridia in massa, sicca, (15–)25–10(–50)  $\times$  3–7  $\mu$ m; conidia quaeque quaque in cellula facilitatem induta nova producere conidia in catenis.

**TYPE SPECIMEN:** CUBA. CIUDAD DE LA HABANA: SANTIAGO DE LAS VEGAS, 22°58' N; 82°20' W, on bark of decaying nuts of *Couroupita guianensis* Aubl. (Lecythidaceae), 7.IV.2000. R.F. Castañeda, (Holotype: INIFAT C00/36–3).

COLONIES on the natural substratum hairy, funiculose, effuse, dark green, olivaceous, or brown. Mycelium superficial and immersed. Hyphae septate, branched, smooth, pale brown, 1–2  $\mu\text{m}$  diam. CONIDIOPHORES micronematous, mononematous, simple or branched, septate, brown or olivaceous, smooth, sometimes reduced to a single conidiogenous cell. CONIDIOGENOUS CELLS monoblastic, terminal, determinate; sometimes polyblastic with sympodial proliferations, 5–10  $\times$  3–6  $\mu\text{m}$ . CONIDIAL SECESSION schizolytic. CONIDIA cylindrical, oblong to bacilliform, (2–)8–9(–16)-septate, olivaceous to pale olivaceous-brown or brown, (15–)25–10(–50)  $\times$  3–7  $\mu\text{m}$ , repeatedly and randomly blastocatenate, forming several irregular chains from several indeterminate cells across the length of the conidial body. Teleomorph unknown.

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## MYCOTAXON

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***Phaeocollybia purpurea* (Cortinariaceae),  
a new species from Wuyishan, China**T.-Z. WEI<sup>1</sup>, S.-Z. FU<sup>1</sup>, P.-P. QU<sup>2</sup> & Y.-J. YAO<sup>1, 3, \*</sup>

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**Abstract** — A new species in *Phaeocollybia*, *P. purpurea*, is described in this paper based on collections from Wuyishan, Fujian Province, China. The new taxon is distinct within the genus for its persistently purple basidiomata, non-viscid pileus, and small basidiospores. The morphological characters used to distinguish the new species from its related species are also provided and discussed in this paper.

**Key words** — taxonomy, morphology, Agaricales, Hymenogastraceae

**Introduction**

*Phaeocollybia* R. Heim is an agaric genus, characterized by its umbonate pileus, subterranean pseudorhiza, brown ornamented basidiospores, and the presence of tibiiform diverticula (Smith 1957, Horak 1977, Redhead & Malloch 1986, Norvell 1998, 2000). The genus is widely distributed in moist temperate (Smith 1957, Horak 1977, Redhead & Malloch 1986, Rees & Wood 1996, Norvell 2000) and tropical zones (Singer 1970, Horak 1980, Horak & Halling 1991, Halling & Horak 2008). However, the ecological status of *Phaeocollybia* still remains uncertain. Smith (1957) inferred that the genus might contain both saprobes and mycorrhiza-formers, while Singer (1986) considered that members of the genus were not obligatorily ectomycorrhizal. Norvell (1998) presented evidence for consideration of *Phaeocollybia* as a mycorrhizal genus.

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*Phaeocollybia* is commonly placed in *Cortinariaceae* (Singer 1986, Kirk et al. 2008, Norvell & Exeter 2009) while Matheny et al. (2006) proposed a molecular-based classification placing the genus in *Hymenogastraceae*, for which further molecular research may provide more evidence. Of the 96 names published in *Phaeocollybia* (CABI 2010), ~50 species are currently accepted by Kirk et al. (2008). *Phaeocollybia* species have been mostly documented from North America and Mexico (Smith 1957, Singer 1970, Smith & Trappe 1972, Horak 1977, Redhead & Malloch 1986, Guzmán et al. 1987, Bandala et al. 1989, 1996, Norvell 2000, 2002, 2004, Norvell & Redhead 2000, Redhead & Norvell 2004, Norvell & Exeter 2007, Halling & Horak 2008), with some from Europe (Pearson 1952, Horak 1977), Asia (Horak 1974, 1977, 1980), South America (Singer 1970, Horak 1977, Horak & Halling 1991), and Oceania (Horak 1973, 1977, Rees & Wood 1996).

The first *Phaeocollybia* species described from China was *P. similis* (Bres.) Singer, based on a collection from Yunnan (Horak 1977). Later, more species were found in China, including a new species, *P. sparsilamellae* P.G. Liu (Liu 1995). Currently, there are 10 species of the genus reported from China (Bi et al. 1994, Deng et al. 2005, Yuan & Sun 1995, Wen et al. 2001, Liu & Qian 2002, Fan 2009).

During a recent expedition to Wuyishan, Fujian Province, China, an undescribed *Phaeocollybia* species was found. A full description of the new taxon is provided in this paper.

### Materials and methods

The fresh basidiomata were photographed after collected from the field in the summer of 2009 and the macro-morphological characters were recorded in detail before drying in an oven at around 45°C. Color names were taken from Ridgway (1912). A 20% KOH solution was used on fresh pileus and stipe surfaces, lamella, and context for chemical reaction. Observation of the reactions was performed under ultraviolet light at a wave length of 360 nm. The specimens are housed in the Mycological Herbarium, Institute of Microbiology, Chinese Academy of Sciences (also as Herbarium Mycologicum Academiae Sinicae, HMAS). Descriptions and line drawings of the micromorphological characters were based on dried collections. Thin sections were prepared by hand with a razor blade. The sections of dried basidiomata were mounted in 5% KOH solution. Basidiospores, basidia, tramal hyphae, context, and cutis of pileus and stipe were measured using an ocular micrometer. At least 30 basidiospores and 20 basidia of each mature collection were measured. The microscopic structures were drawn with the aid of a camera lucida.



FIG. 1. Basidiomata of *Phaeocollybia purpurea* (HMAS 250001, holotype).

### Taxonomy

*Phaeocollybia purpurea* T.Z. Wei, S.Z. Fu, P.P. Qu & Y.J. Yao, sp. nov. FIGS 1–2.

MYCOBANK MB 518112

*Pileus* 2.0–6.0 cm *latus*, *primo* conicus, *dein* umbonato-applanatus, *superficie* *primo* obscuro-violaceus, *atro-griseo-purpureus* vel *brunneo-purpureus*, *dein* brunneus vel *purpurate ferrugineo-brunneus*, glaber, sericeus, nonviscidus. *Lamellae* adnatae, confertae, ad 4 mm latae, *primo* pallide violaceo-griseae, *dein* violaceo-griseae vel griseo-violaceae, *demum* brunneo-purpureae *interdum* maturitate ferruginescenter suffusae. *Stipes* centralis, 2.0–5.5 × 0.3–0.6 cm, cylindraceus, *superficie* atro-purpureo-griseus vel atro-griseo-violaceus, glaber, sericeus, fistulosus. *Pseudorhiza* ad 8.0 cm longa, concolor. *Basidiosporae* 3.5–5.0 × 3.0–4.0 µm, ovoideae vel ellipsoideae, brunneolae vel ferrugineo-brunneae, leviter verrucosae. *Basidia* 18.0–24 × 5.0–6.0 µm, clavata, tetraspora.

TYPE—CHINA, Fujian Province, Wuyishan National Nature Reserve, in broad leaved forest, 27°44.935'N, 117°40.652'E, alt. 715 m, 18 June 2009, T.-Z. Wei 300, Holotype HMAS 250001; Isotype K(M) 166177.

ETYMOLOGY—*purpurea*, from the Latin for 'purple', referring to the color of the basidiomata.

BASIDIOMATA scattered to gregarious. PILEUS 2.0–6.0 cm diam., conical at first, then convex, finally expanding to applanate with a bluntly pointed umbo; margin decurrent at first and then straight; surface dull violet (Dusky violet), dark grayish purple (Dull Dusky Purple) to brownish purple (Deep Livid Purple) when young, changing to brown (Liver Brown) or rust-brown (Hay's Russet) with distinct purplish tint, center more or less darker; smooth, glabrous, silky, hygrophanous, neither viscid nor lubricous when moist, opaque. CONTEXT up to 4.0 mm thick at center, fleshy, purple close to pileus cuticle, elsewhere pale gray with purplish tint. LAMELLAE adnate, up to 4.0 mm broad, ventricose, crowded,

with lamellulae; pale violet gray (Pale Violet Gray) when young, then violet-gray (Deep Violet Gray) to grayish violet (Dark Grayish Blue-Violet), brownish purple (Dark Livid Purple) with rust tint when mature; edge pallid, uneven in age. STIPE central, above-ground part  $2.0\text{--}5.5 \times 0.3\text{--}0.6$  cm, cylindrical, with subterranean pseudorhiza; surface dark purplish gray (Vinaceous-Slate) to dark grayish violet (Dark Grayish Blue-Violet), occasionally tinted rust from basidiospores, smooth, glabrous, silky; hollow, with concolorous cortex; cortex  $1.0\text{--}1.5$  mm thick, brittle, cartilaginous. PSEUDORHIZA up to 8.0 cm long, rhizomorphic, cylindrical and hollow above, tapering below and solid, concolorous with stipe surface or slightly darker; cortex  $1.5\text{--}2.0$  mm thick, cartilaginous. VEIL REMNANTS not observed. BASIDIOSPORE DEPOSIT brownish rusty.

BASIDIOSPORES  $3.5\text{--}5.0 \times 3.0\text{--}4.0$   $\mu\text{m}$ , ellipsoid ovoid to ellipsoid with eccentric apiculus and bluntly round to pointed apical callus in profile, brownish to rusty brown, finely punctate to verruculose, thick-walled, inamyloid. BASIDIA  $18.0\text{--}24 \times 5.0\text{--}6.0$   $\mu\text{m}$ , clavate, 4-spored, with long (up to 6  $\mu\text{m}$ ) sterigmata, hyaline to subhyaline, thin-walled, basally clamped. LAMELLA EDGE heterogeneous, crowded with abundant cheilocystidia and few basidia. CHEILOCYSTIDIA abundant,  $20\text{--}29(-40) \times (2.3\text{--})3.5\text{--}5.0$   $\mu\text{m}$ , clavate to ampullaceous, usually with a mucronato-capitate apex atop a short narrow refractive neck, hyaline, thin-walled. PLEUROCYSTIDIA none found. HYMENOPHORAL TRAMA  $60\text{--}120$   $\mu\text{m}$  wide, regular, of thin-walled hyphae; hyphae  $4.0\text{--}12.0$   $\mu\text{m}$  diam., hyaline, rare yellowish, thin-walled. SUBHYMENIAL LAYER  $3.0\text{--}6.0$   $\mu\text{m}$  wide, of repent branched hyphae; hyphae  $2.0\text{--}3.5$   $\mu\text{m}$  diam, thin-walled, hyaline to subhyaline. PILEIPELLIS bilamellate, compact, yellowish brown, of thick-walled gelatinized hyphae gel-encrusted with yellow-brown pigments often concentrating at septa; suprapellis  $20\text{--}50$   $\mu\text{m}$  wide, hyphae  $2.5\text{--}5.0$   $\mu\text{m}$  diam.; subpellis  $80\text{--}150$   $\mu\text{m}$  wide, hyphae  $5.0\text{--}15.0$   $\mu\text{m}$  diam, most elements thick-walled. PILEAL TRAMA of branched hyphae, hyphae normally  $4.0\text{--}8.0$   $\mu\text{m}$  diam., sometimes inflating to  $16$   $\mu\text{m}$  diam., thin-walled, hyaline to brownish, rarely with purplish content when observed in water. STIPITPELLIS of longitudinally parallel hyphae, hyphae  $1.5\text{--}4.0$   $\mu\text{m}$  diam., thick-walled, pale brown. STIPE TRAMA bilamellate, vessel hyphae longitudinally parallel, hyphae  $5.0\text{--}20$   $\mu\text{m}$  diam., thick-walled (up to  $3$   $\mu\text{m}$  wide); inner surface of longitudinally subparallel hyphae, hyphae  $1.5\text{--}4.0$   $\mu\text{m}$  diam., thin-walled, hyaline, rare subhyaline. PSEUDORHIZA strongly sarcodimitic with thick-walled vessel hyphae predominant. TIBIFORM DIVERTICULA thin-walled, hyaline, up to  $12.0$   $\mu\text{m}$  long,  $0.5\text{--}1.0$   $\mu\text{m}$  diam., abundant on pseudorhizal pellis and basal mycelium, subcylindrical and with globose apex, with no septum between base and hypha. CLAMP CONNECTIONS abundant in stipe trama, less frequent but present at basidial bases, cheilocystidia, pileipellis, and stipitipellis.

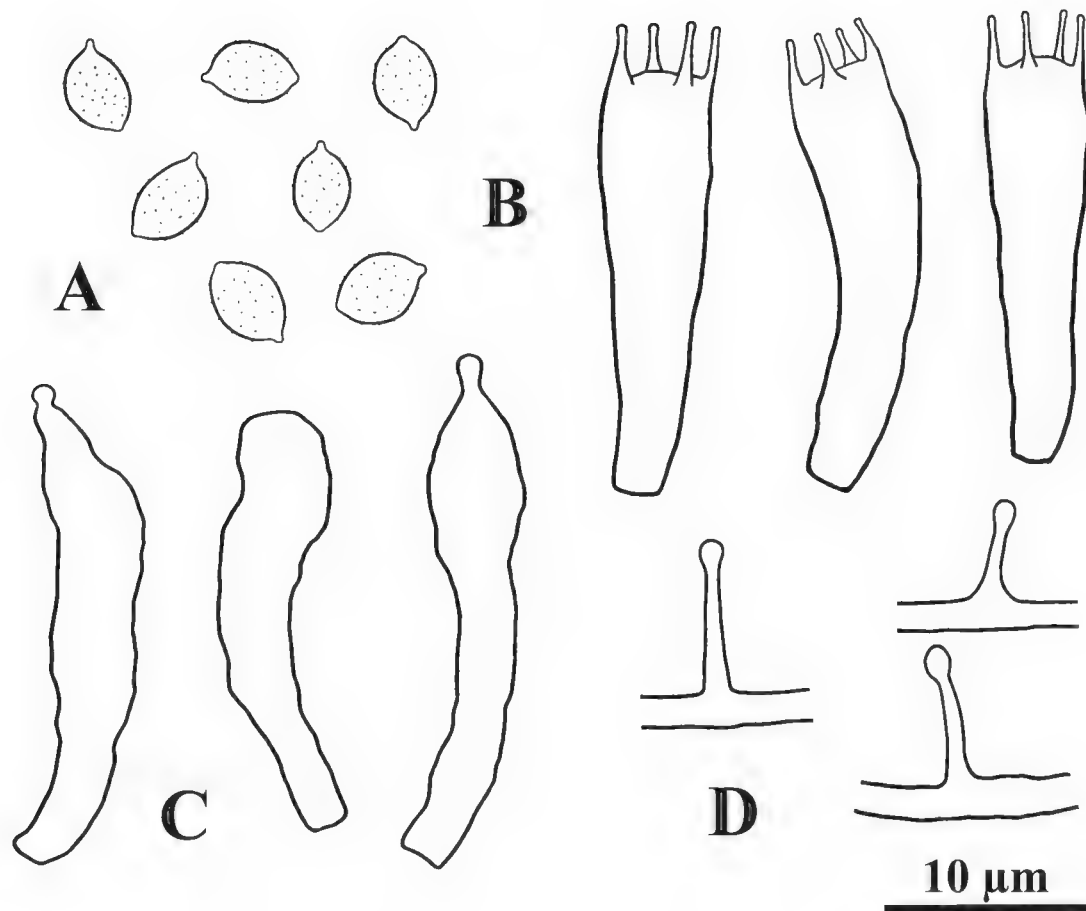


FIG. 2. *Phaeocollybia purpurea* (HMAS 250001, holotype).  
A. basidiospores; B. basidia; C. cheilocystidia; D. tibiiform diverticula.

CHEMICAL REACTION with 20% KOH blackening at all parts. FLUORESCENCE REACTION UNDER ULTRAVIOLET LIGHT bright yellow in lamellae, pale yellow with purplish tint in context, violet-purple in all other parts. TASTE of context mild. ODOR of context indistinct.

HABITAT—on ground in forest dominated by broadleaf species and mixed with a few conifers, near *Quercus* sp. and *Cunninghamia* sp.

ADDITIONAL SPECIMEN EXAMINED – CHINA, Fujian Province, Wuyishan National Nature Reserve, in broad-leaved forest, 27°44.935'N, 117°40.652'E, alt. 715 m, 22 June 2009, Wei T.-Z. 595, HMAS 250002.

The two collections examined here are placed in *Phaeocollybia* based on their pileal umbo, cartilaginous stipe, long pseudorhiza, brown verrucose basidiospores, and tibiiform diverticula. The newly described *P. purpurea* is mainly distinguished from other taxa in the genus by its violet to purple basidiomata. Four other *Phaeocollybia* species — *P. amygdalospora* Bandala & E. Horak (Bandala et al. 1996), *P. parvispora* Corner & E. Horak (Horak 1977), *P. pseudolugubris* Bandala & E. Horak (Bandala et al. 1996), *P. singularis* E. Horak & Halling (Horak & Halling 1991) — produce similar basidiomata

with lilac, purple or violaceous tints all-over when young. However, the violet to purple colors persist in the new species whereas the violet tones are lost over time in the four species cited above. Further, *P. purpurea* is separated from *P. amygdalospora*, *P. pseudolugubris*, and *P. singularis* by its non-viscid pileus when moist and its smooth glabrous pileus lacks the appressed squamules typical of *P. parvispora*.

Microscopically, its small basidiospore size ( $3.0\text{--}5.0 \times 3.0\text{--}4.0 \mu\text{m}$ ) clearly separates *P. purpurea* from *P. amygdalospora* ( $6.0\text{--}9.0 \times 4.0\text{--}5.0 \mu\text{m}$ , Bandala et al. 1996), *P. pseudolugubris* ( $8.0\text{--}10.0 \times 4.0\text{--}5.0 \mu\text{m}$ , Bandala et al. 1996), and *P. singularis* ( $8.0\text{--}9.5 \times 4.5\text{--}5.0 \mu\text{m}$ , Horak & Halling 1991), all of which have amygdaliform to limoniform shaped spores. *Phaeocollybia parvispora* also has small ellipsoid basidiospores ( $3.4\text{--}4.5 \times 2.5\text{--}3.0 \mu\text{m}$ , Horak 1977), but they are considerably narrower than those of the new species.

Two other species, *P. arduennensis* Bon and *P. bicolor* E. Horak, produce similar cheilocystidia, clamps, and small spores. In addition, the brown pileus of *P. arduennensis* has a purplish tinge, and the lamellae of *P. bicolor* are lilac at first. However, the ochreous orange lamellae of *P. arduennensis* (Bon 1992) contrast with the violet to brownish purple lamellae in *P. purpurea*, while *P. bicolor* is distinguished by an avellaneous to light brown pileus and the absence of a pseudorhiza (Horak 1977).

DNA sequences derived from the nuclear ribosomal DNA internally transcribed spacer region (nrDNA-ITS) from our laboratory specimens and compared with sequences now on deposit in Genbank support *P. purpurea* as a distinct species in *Phaeocollybia* that clusters with *P. ratticauda* E. Horak (AF501568.1: voucher BRV 99/11) in the same terminal clade but with relatively long branches (unpublished data). *Phaeocollybia ratticauda* resembles *P. purpurea* in lilac coloration (lamellae and stipe) and small basidiospores ( $5.0\text{--}6.0 \times 3.5\text{--}4.0 \mu\text{m}$ ). However, its dark brown to liver brown pileus (Horak 1973) differentiates *P. ratticauda* from *P. purpurea*. The results of our DNA sequence analyses of *Phaeocollybia* species will be published elsewhere.

### Acknowledgements

Dr. Lorelei L. Norvell and Prof. P.-G. Liu are acknowledged for serving as pre-submission reviewers and for their valuable comments and suggestions. The authors are grateful to Prof. J.-Y. Zhuang for his help in correcting the Latin description of the new taxon and critical review of the manuscript. This project is supported by the Chinese National Science & Technology Project (2008BADA1B01) and the Innovation Project of the Chinese Academy of Sciences (KSCX2-YW-G-074-04).



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## MYCOTAXON

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**Contributions to the lichen flora of the  
Hengduan Mountains, China 1.  
Genus *Pseudephebe* (lichenized Ascomycota, Parmeliaceae)**

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**Abstract** — *Pseudephebe pubescens* is reported, described and illustrated from the Chinese Hengduan Mountains region. It is characterized by its slender and isotomic-dichotomous branched filaments forming tiny cushions, a cortex of longitudinally oriented hyphae that become prosoplectenchymatous at the surface, medullary hyphae that are not ornamented, and the absence of lichen substances. It grows on arctic-alpine rock.

**Key words** — alpine lichens, Sichuan, taxonomy, thallus anatomy, Xizang

## Introduction

The Hengduan Mountains are part of the Himalayas located in southern China, including western Sichuan, northwestern Yunnan, and southern Xizang (Tibet). This mountain range has an area of ca. 364,000 km<sup>2</sup>. The region has one of the highest biodiversities of lichens in the world. In recent decades, many interesting lichens were found in the region (McCune et al. 2003, Jørgensen 2003, Obermayer 1997, 2001, 2003, 2004; Wang et al. 2001, 2003, 2005; Xiao et al. 2006, Niu et al. 2007, 2008). Although we began our taxonomic work under the series entitled “Taxonomic study on the lichen genus *Bryoria* (lichenized Ascomycota, Parmeliaceae) from the Sino-Himalayas” (Wang et al. 2006), we prefer to title the new series as in the present paper, because the Hengduan Mountains form a more distinct biogeographical region than the Sino-Himalayas. In this paper, the genus *Pseudephebe* is reported from the Hengduan Mountains as new for China.

## Materials and methods

The four specimens were collected in W-Sichuan and S-Xizang between 2001 and 2007. The collections were annotated and photographed in the field. Descriptions of external morphology were based on air-dried materials observed under a dissecting stereomicroscope. Sections were made with a razor blade under the stereomicroscope, and mounted in GAW (glycerol: ethanol: water=1:1:1). SEM micrographs were obtained with the scanning electron microscope JEOL JSM-5410 LV of National Instrumentation Center for Environmental Management, Seoul National University. Thin-layer chromatography (TLC) was performed to identify lichen chemical compounds with three developing solvent systems (Culberson 1972). The specimens used in this study are deposited in the Cryptogamic Herbarium, Kunming Institute of Botany, Academia Sinica (KUN).

## Taxonomy

*Pseudephebe* M. Choisy, Icon. Lich. Univ. ser. 2, fasc. 1: [sine pag.] (1930).

TYPE SPECIES: *Pseudephebe pubescens*

Thallus fruticose, appressed to the substrate; branching isotomic-dichotomous, the branches terete but tending to become dorsiventrally compressed in one species, even or uneven, brown to black, dull to shiny; cortex of longitudinally oriented hyphae which become prosoplectenchymatous at the surface; medullary hyphae not ornamented. Apothecia lateral; thalloid margin concolorous with the thallus, sometimes ciliate, asci clavate; spores 8, ellipsoid, hyaline, simple, 7–12 x 6–8  $\mu\text{m}$ . Pycnidia common. Lichen products absent.

Only two species of this genus are known worldwide (Brodo & Hawksworth 1977, Nash et al 2002: 409–411).

*Pseudephebe pubescens* (L.) M. Choisy, Icon. Lich. Univ., ser. 2, 1: [sine pag.] (1930).

FIGS. 1–3

= *Lichen pubescens* L., Sp. Pl. 2: 1155 (1753).

= *Alectoria pubescens* (L.) R. Howe, Classif. Usneac. Amer.: 23 (1912).

Thallus fruticose, decumbent to subpendulous, forming small cushions, loosely adnate, more or less circular c. 2–12 cm diam., brown to blackish brown, smooth, dull to slightly shining; main branches slender, cylindrical, uneven, 0.1–0.2 mm diam., 0.05–0.1 mm near the tips; branching frequent, isotomic-dichotomous, not flattened; sometimes with circular pits presented on the surface (FIG. 2); true lateral spinules, soredia, isidia and pseudocyphellae absent; Cortex 50  $\mu\text{m}$  thick, 2-layered, with rectangular to irregular and knobby cells at the surface; medulla white, medullary hyphae not ornamented (FIG. 3). Apothecia not seen. Pycnidia common on tubercles, especially near the axils



FIG. 1. Habit of *Pseudephebe pubescens* in its natural habitat in Sichuan, China (photograph by Wang, 5 June 2006, voucher: Wang Li-song 06-26090).

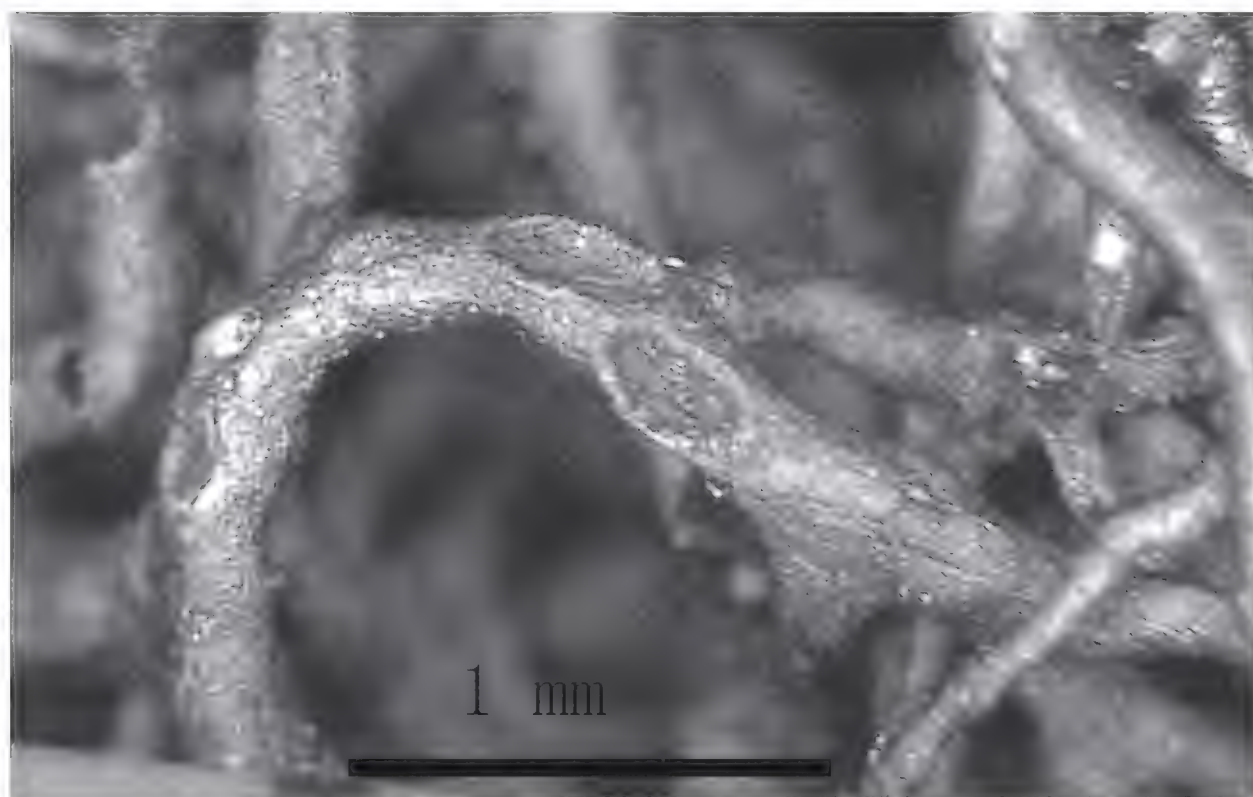


FIG. 2. Close-up of the thallus of *Pseudephebe pubescens*, showing uneven main branch sometimes with circular pits on the surface of the cortex (under the dissecting stereomicroscope; Wang Li-song 06-26090).



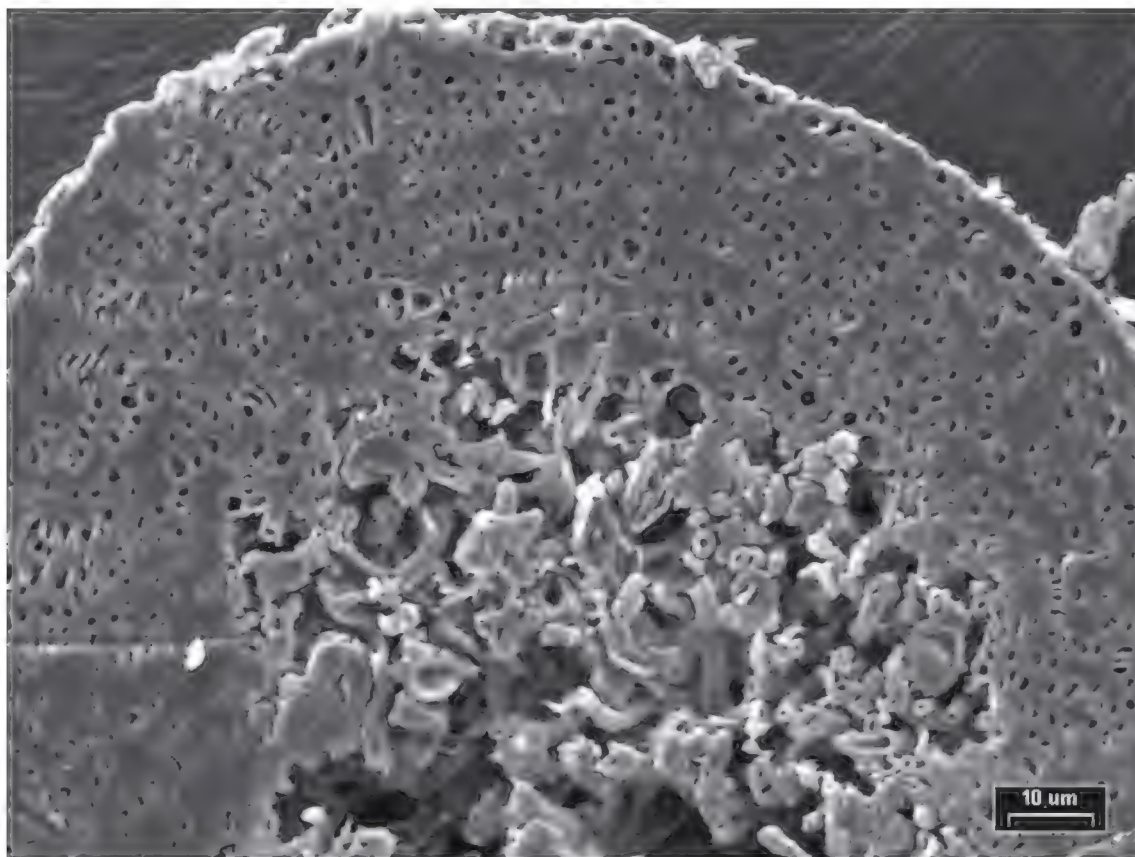


FIG. 3. Cross section of the main branch of *Pseudephebe pubescens* under SEM (Wang Li-song 83-2152a).

and bases, dark brown to black, 0.5–0.7 mm diam., conidia bifusiform, 6–7.5 x 1–2  $\mu$ m, colorless. Photobiont: green algae. Cortex and medulla: K–, C–, KC–, P–; no lichen substances detected by TLC.

**HABITAT AND ECOLOGY** —Thallus loosely attached on siliceous rock surfaces, forming small cushions (FIG.1). All collections were from the alpine zone, between 4330 and 5070 m elevations in the Hengduan Mountains. Associated species included *Umbilicaria indica*, *Ophioparma ventosa*, *Rhizoplaca chrysouleuca* and *Rhizocarpon* spp.

**DISTRIBUTION** —*Pseudephebe pubescens* is widely distributed. It is found in Europe (Hawksworth 1972), North America (Brodo & Hawksworth 1977), California (Nash et al. 2002: 409–411), the Alaskan arctic slope (Thomson 1979: 240–241), Japan (Kurokawa et al. 1968, 1981), and the southern hemisphere (Brodo & Hawksworth 1977). It is new to China (FIG. 4).

**SPECIMENS EXAMINED**—CHINA. Sichuan Prov., Kingding County, Zheduoshan Mt., 30° 04' N, 101° 48' E, 4200–4330 m, on rock, Wang Li-song 06-26090, 07-29009; Muli County, Sanqu village, 4400 m, on rock, Wang Li-song 83-2152a; Xizang (Tibet) Prov., Naidong County, 28° 37' N, 92° 13' E, 5070 m, on rock, Wang Li-song 07-28595 (FIG. 4). Additional specimens examined: numerous North American specimens and Finland: Lapponia enontekiensis(le). Enontekio, Hetta, Jyppyra, ad rupem in summon monte. 21 VII 1957, Coll. A. J. Huuskonen (no number).

COMMENTS —The main branches are wider (0.2–0.5 mm diam.) in specimens from the Alaskan arctic slope (Thomson 1979: 240–241) than in the Chinese materials, where they are only 0.2 mm in diam.

This species differs from *Pseudephebe minuscula* (Nyl. ex Arnold) Brodo & D. Hawksw. in that the latter has somewhat flattened branches and shorter internodes (Brodo & Hawksworth 1977). The two species do, however, tend to integrate. Although the Chinese specimens are somewhat smaller and have shorter internodes than typical *P. pubescens*, the absence of distinctly flattened branches indicates *P. pubescens*.

In North America *P. minuscula* tends to have a more continental distribution than *P. pubescens* (Brodo & Hawksworth 1977). Although the climate of the Hengduan Mountains has no close analog in North America, the Hengduan Mountain region is influenced by both continental and maritime air masses, offering a wide range of habitats for alpine lichens. The abundance of cyanolichens at lower elevations in some parts of the Hengduan Mountains suggests a suboceanic climate.

The genus *Pseudephebe* is close to the genus *Bryoria* Brodo & D. Hawksw., which also contains fruticose alpine species found on rocks from the Hengduan Mountain region. For example, *Bryoria nitidula* and *B. tenuis* are similar to *Pseudephebe* in having a dark brown to blackish thallus and medullary hyphae that are not ornamented. However, the alpine *Bryoria* species on rock are

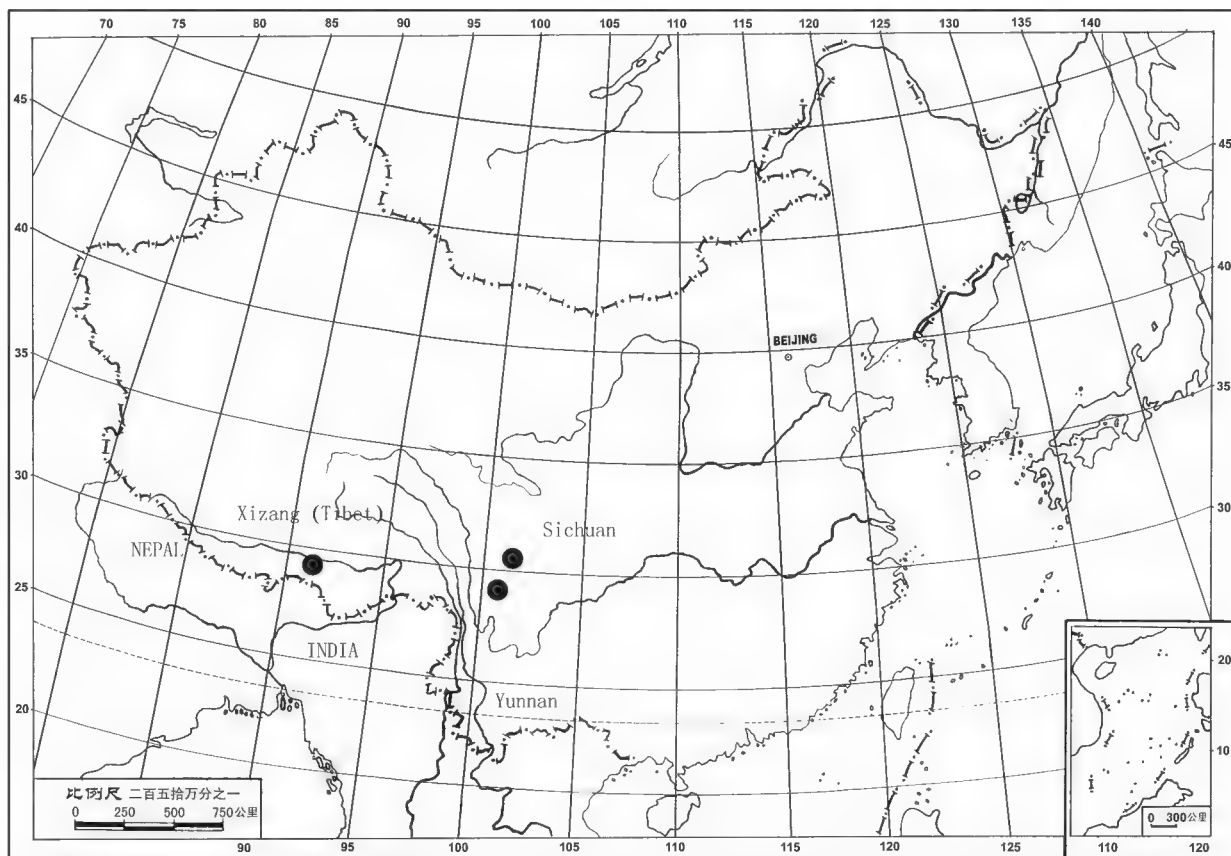


FIG. 4. Distribution of *Pseudephebe pubescens* in China.

usually larger (branches >3 cm long), have an erect to caespitose thallus and often have pseudocyphellae. Furthermore, they usually contain the substances atranorin or fumarprotocetraric acid.

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## MYCOTAXON

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**Myxomycete diversity from arid and semiarid zones  
of the Canary Islands (Spain)**E. BELTRÁN-TEJERA<sup>1\*</sup>, J. MOSQUERA<sup>1</sup> & C. LADO<sup>2</sup>

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**Abstract** — A study of the myxomycetes recovered from the arid, semiarid, and dry zones of the Canary Islands is presented herein. A total of sixty-three species, most growing on succulent plants, is reported. *Physarum bethelii*, *P. confertum*, and *Stemonitis herbatica* are cited for the first time from the Canaries, with additional new records from each island. The importance of the endemic plants such as *Euphorbia canariensis* as substrates for myxomycetes is analyzed. As reported from other arid zones of the world species belonging to the orders *Physariales* and *Trichiales* dominate. *Badhamia melanospora*, commonly recorded from the deserts of America, was the most frequently recovered species from the Canaries. The parallel between the myxobiotas of the dry areas of the Americas and the Canary Islands is also discussed. The complete study and catalogue is available at <http://www.mycotaxon.com/resources/weblist.html>.

**Key words** — biodiversity inventory, Macaronesian bioregion, thermophilous habitats, xerophytic substrates

**Introduction**

The study of myxomycetes from the arid lands of the world is a subject of recent interest (Lado et al. 1999, 2009; Mosquera et al. 2000a,b, 2003; Wrigley de Basanta et al. 2008, 2009). Cacti and other succulent plants have been found to support a characteristic succulenticolous species assemblage (Lado et al. 1999). Inventories of the myxomycetes from some deserts, especially from the Americas and other regions of the world, have been published in the last decades (Blackwell & Gilbertson 1980; Novozhilov et al. 2006; Lado et al. 2007a,b; Estrada-Torres et al. 2009) but information about the myxomycetes of insular arid lands is very scarce (Eliasson 1971, 1991, 2004).

The Canary Islands are a group of islands of volcanic origin located in the Atlantic Ocean, between 27°40'–29°30'N latitude and 13°25'–18°10'W

longitude, approximately 100–500 km from the west African coast and the Sahara desert and on about the same latitude as Florida (USA). The Archipelago is composed of seven major islands (Hierro, La Palma, Gomera, Tenerife, Gran Canaria, Fuerteventura, Lanzarote) and a few smaller ones. Due to their volcanic nature the relief is very abrupt, and the elevation ranges from sea level to 3718 m, on Tenerife Island. The considerable elevation gradient produces substantial environmental variation with respect to temperature and moisture across the islands.

The vegetation of the Canary Islands is highly stratified due to the influence of climatic factors, altitude, and exposure. From a bioclimatic point of view, there are six ombrotypes in the Canaries (hyperarid, arid, semiarid, dry, subhumid, humid). From sea level to 200–400 m on their northern slopes and up to 600–1000 m on the southern side, there is an arid-semiarid-dry climate, characterized by high temperatures (18°–22°C) and low annual precipitation (50–350 mm). The vegetation of these zones represents a characteristic xerophytic scrubland (called “cardonal-tabaibal” in Spanish) with succulent plants and occasional aphyllous or spiny shrubs dominated by *Euphorbia* spp. with a high proportion of endemic plants (> 50%). Above the *Euphorbia* communities there are woodland and forest belts, followed by dry xerophytic summit vegetation represented only in the highest islands.

In several places the natural vegetation was replaced by cultivated plots, and many exotic species such as *Opuntia* spp. and *Agave* spp., were introduced. Presently these disturbed formations form part of the Canary landscape. These are the anthropic plant communities.

The overall aim of our investigation was to study the myxomycetes associated with arid, semiarid, and dry zones. As a result, most of the sampled stations were located in the lower elevations of the islands, between sea level and 500 m.

### Material and methods

During eleven years (1994–2005) 72 localities were sampled at lower elevations (generally below 500 m) across seven of the Canary Islands. Microscopic measurements were made from material directly mounted in Hoyer's medium. An Olympus BH-2 and a Zeiss Jenemad-2 achromatic phase contrast microscope were used in the identification of the specimens. The specimens have been deposited in TFC Mic, and MA-Fungi herbaria. Nomenclature largely follows that of Lado (2001).

### Results

A total of 63 species of myxomycetes were recovered, of which *Physarum bethelii* T.Macbr. ex G.Lister, *P. confertum* T.Macbr., and *Stemonitis herbatica*

Peck are reported for the first time from the Canaries. The taxa recovered were distributed across 21 genera, among which *Physarum* has the greatest representation with 15 species, followed by *Didymium* (12 species), and *Arcyria* (7). As reported from other arid zones of the world the species belonging to the order *Physarales* and *Trichiales* dominate. *Badhamia melanospora*, commonly recorded from the deserts of America, was the most frequently recovered species from the Canaries.

The analysis of the substrates was based on a total of 463 samples, collected from 34 vascular plant species, of which 14 are characterized by succulent biotypes, 19 are woody, and 1 herbaceous. The greatest number of myxomycete species (51) was collected from succulent plants. Of these 32 could be characterized as strictly succulenticolous, since they were only observed from this type of substrate, whereas the remaining species appeared on woody remains and/or leaf litter. *Euphorbia canariensis* was found to be the most productive substrate with respect to species richness among the endemic succulent species with a total of 117 collections distributed across 27 species of myxomycetes. *Opuntia maxima* was the most productive substrate among the introduced succulent species, with 138 samples belonging to 23 species of myxomycetes.

This study was carried out in the same way as research on the myxomycetes of arid lands in Mexico (Estrada-Torres et al. 2009). Some of the results have been similar and have resulted in several taxa new to science (e.g. *Cribraria zonatispora*, *Trichia agaves*, *Licea succulenticola*, and *Didymium wildpretii*) having been described based on material from both areas. Several centuries ago succulent plants and cacti from America were introduced to the Canary Islands to see whether they could become acclimated and be cultivated in Europe. Therefore, the similarity in the myxobiota of these areas could potentially have been influenced, as has been suggested previously (Lado et al. 2007b), by the introduction of these plants.

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## MYCOTAXON

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**A new species of *Ijuhya*, *I. antillana*,  
from the French West Indies**CHRISTIAN LECHAT<sup>1\*</sup> & RÉGIS COURTECUISSE<sup>2</sup><sup>1</sup>\*lechat@ascofrance.fr

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**Abstract** — A detailed description of *Ijuhya antillana* sp. nov. is presented based on two collections on dead inflorescences of *Heliconia caribaea* in Guadeloupe and Martinique. The *Acremonium*-like anamorph has been obtained in culture. A key to the species of *Ijuhya* with fasciculate hairs is presented.

**Key words** — *Ascomycota*, *Bionectriaceae*, *Heliconiaceae*

**Introduction**

During the course of a research program on the fungal diversity of Lesser Antilles, conducted by Prof. R. Courtecuisse under the auspices of Société Mycologique de France with the funding from ONF (French Forest Office) and DREAL (Martinique delegation of French Environment ministry), interesting collections of *Hypocreales* have been made in different localities and ecological situations in Martinique and Guadeloupe. A species first collected in August 2007 in Martinique represents a new taxon of the genus *Ijuhya*. Later, a second specimen of the same species from Guadeloupe was cultured from single ascospores that produced an asexual state *Acremonium*-like. A description and illustrations of this new species are presented here.

**Materials & methods**

Specimens were examined using the methods described by Rossman et al, (1999). Microscopic observations and measurements were made in water and ascospore ornamentation was observed in lactic cotton blue.



## Taxonomy

*Ijuhya antillana* Lechat & Courtec., sp. nov.

FIGS. 1

MYCOBANK MB 516744

*Ascomata subglobosa, apice applanata, 160–230 µm diametro, subhyalina vel aurantia, corona subapicalis pilis agglutinatis hyalinis vel aurantia, crasse-tunicatis, flexuosis composita, colore in KOH non mutanda. Asci 60–75 × 6.5 µm, octospori, unitunicati, inamyloidei. Ascospores fusiformes vel ellipsoideae, (10.5–)11–13(–14) × 2.5–3.5 µm, uniseptatae, sublaeves. Status asexualis: Acremonii similis*

**HOLOTYPE:** French West Indies, Martinique, Morne Rouge, la Propreté, 29 Aug. 2007, on dead inflorescence of *Heliconia caribaea* (*Heliconiaceae*), Christian Lechat CLL7321 (LIP); ex-type culture CBS 122797.

**ETYMOLOGY:** The epithet refers to The Lesser Antilles, the region where this species was collected twice.

**ASCOMATA** gregarious, solitary or crowded in groups of 2–3, superficial, subglobose, apex flattened with a minute papilla, 120–160 µm high × 160–230 µm diam ( $m = 145 \times 210 \mu\text{m}$ ,  $n = 15$ ), white when immature, later dark orange to brownish-orange, collapsing cupulate when dry, not changing colour in 3–5% KOH or lactic acid. Perithecial wall abundantly covered by flexuous hyphae 2.5–3 µm diam developing from ascomatal base, apex surrounded by thick-walled hairs except on papilla, hairs 100–160 µm long, 2.5–3 µm wide, pale yellowish to brownish-orange when dry, cylindrical, slightly flexuous, thick-walled, wall 0.7–1 µm thick, rounded at tip, septate, arising from cells of ascomatal wall, fasciculate, agglutinated into triangular teeth 100–160 µm long × 20–30 µm wide at base, arranged in a stellate fringe around upper margin of perithecia.

**PERITHECIAL WALL** 20–30 µm thick, composed of two regions: outer region 12–20 µm wide, of 3–5 layers of globose to elongate cells 3–10 × 3–4.5 µm with yellow wall; inner region 6–10 µm wide, of elongate, flattened, hyaline cells 5–10 × 1.5–3 µm.

**ASCI** 60–75 × 6–8.5 µm ( $m = 71.5 \times 8 \mu\text{m}$ ,  $n = 20$ ), clavate to fusoid, apex flattened, without ring, with 8 obliquely uniseriate or irregularly biseriate ascospores. No interthecial elements seen.

**ASCOSPORES** (10.5–)11–13(–14) × 2.5–3.5 µm ( $m = 12.7 \times 3.2 \mu\text{m}$ ,  $n = 30$ ), hyaline, fusoid-ellipsoidal, straight, equally 2-celled, not constricted at septum, punctate-striate with 2 drops in each cell.

**ANAMORPH:** *Acremonium*-like

**CULTURAL CHARACTERISTICS:** After one week at 25°C on Difco PDA containing 5 mg/L streptomycin, colony 3–4 cm diam, mycelium white, producing an abundant *Acremonium*-like culture in center of colony, composed of

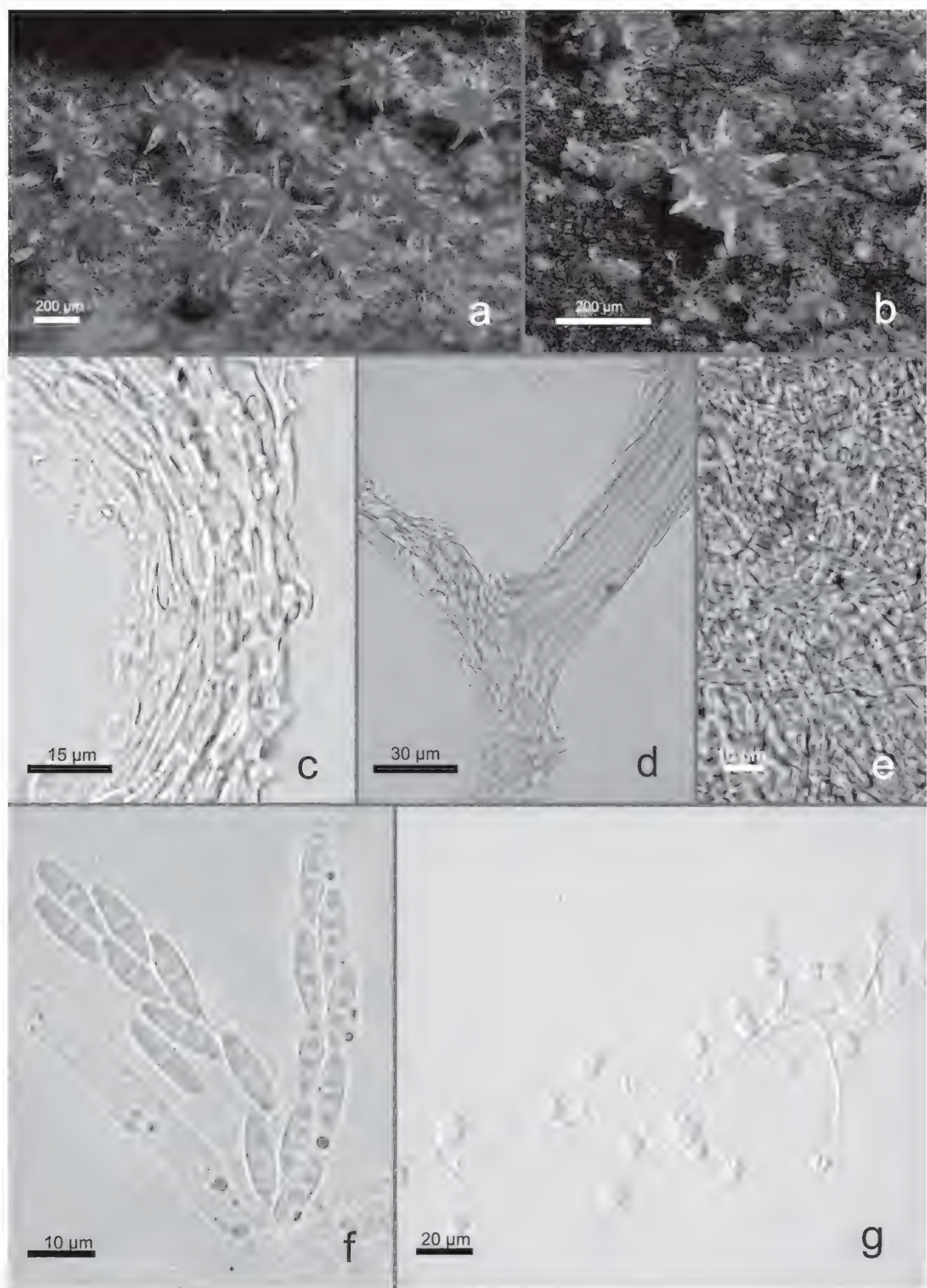


FIG. 1. *Ijuhya antillana*. a. Ascomata b. Single ascoma. c. Median section of perithecium. d. Fasciculate hairs. e. Hyphae covering ascomatal surface. f. Asci. g. *Acremonium*-like anamorph in culture. Additional photos at <http://www.ascofrance.fr>.

monophialidic conidiophores, 28–60 µm long, 2–3 µm diam, arising from smooth hyphae 2–3 µm diam, producing ellipsoidal conidia (4.5–)5–7(–8) × 1.8–3 µm ( $m = 6.4 \times 2.4 \mu\text{m}$ ,  $n = 30$ ), hyaline, smooth, non-septate, with a basal abscission scar.

ADDITIONAL SPECIMEN EXAMINED: French West Indies, Guadeloupe, Petit Bourg, sentier de la Chute de Moreau, 14 Aug. 2008, on dead inflorescence of *Heliconia caribaea*, Christian Lechat CLL8321 (LIP).

Discussion

*Ijuhya antillana* is placed in the genus *Ijuhya* Starbäck based on the ascomata not changing color in 3% KOH or lactic acid, fasciculate hairs around the perithecial apex, striate ascospores, ascomatal wall of small, thick-walled cells and *Acremonium*-like anamorphs as defined by Rossman et al. (1999).

This species is related to several known species of *Ijuhya*, which have a stellate crown of fasciculate, agglutinated hairs around the perithecial apex, such as *I. chilensis* (Speg.) Rossman & Samuels (Rossman et al. 1999), *I. dentifera* (Samuels) Rossman & Samuels (Samuels 1976 as *Nectria dentifera*), *I. equiseti-hiemalis* Lechat & Baral (Lechat & Baral 2008), *I. peristomialis* (Berk. & Broome.) Rossman & Samuels (Rossman et al. 1999), and *I. parilis* (Syd.) Rossman & Samuels (Samuels 1988). The new species differs from these in size and ornamentation of ascospores and/or length of fasciculate hairs.

Key to species of *Ijuhya* with fasciculate hairs

- 1. Hairs 200–300 µm long; ascospores (24–) 30–60(–110) × 4–7(–8) µm, striate; ascomata pale yellow ..... *I. peristomialis*
- 1. Hairs averaging less than 200 µm long ..... 2
- 2. Ascospores averaging less than 12 µm long ..... 3
- 2. Ascospores averaging more than 12 µm long ..... 4
- 3. Ascospores (8.5–)9.5–11.5(–12.5) × 2.8–3.2(–3.5) µm, striate; ascomata brownish-orange, hairs 28–80 × 2–2.5(–3) µm ..... *I. equiseti-hiemalis*
- 3. Ascospores 6–8(–9) × 3–4 µm, spinulose; ascomata orange-yellow, hairs 150–200 × 3–4 µm ..... *I. dentifera*
- 4. Ascospores striate ..... 5
- 4. Ascospores spinulose 14.5–20 × (2.5–)3–5(–5.4) µm, ascomata brownish-orange, hairs 30–50 µm long ..... *I. parilis*
- 5. Ascospores (10.5–)11–13(–14) × 2.5–3.5 µm; ascomata dark orange, hairs 100–160 µm × 2.5–3 µm ..... *I. antillana*
- 5. Ascospores (19–)21–28 × 3.5–4.5 µm; ascomata dull orange, hairs up to 100 µm long ..... *I. chilensis*

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**Status of some poorly known lichen species from the genus  
*Lecanora* (lichenized Ascomycota) in Poland**PAWEŁ CZARNOTA<sup>1, 2\*</sup>, PIOTR OSYCZKA<sup>3</sup> & AGNIESZKA KOWALEWSKA<sup>4</sup>

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**Abstract** — Taxonomic and chorological notes on four *Lecanora* species, misidentified or poorly known in Poland, are presented. *L. aitema* is reported from Poland for the first time; its status and previous reports in the country are discussed. *L. phaeostigma*, practically known only from historical publications, appears to be quite frequent in the Polish Carpathians. The current status of poorly known *L. ramulicola* is presented here based on revised literature and herbarium data. No specimen of *L. cadubriae* has been confirmed in Polish collections and all reports of this species from Poland, in fact, refer to *L. ramulicola*. Because of the misidentifications and nomenclatural confusion, *L. cadubriae* should be excluded from the list of Polish lichens. The taxonomy, nomenclatural remarks, chemistry, habitat requirements, and distribution of all taxa are discussed.

**Key words** — lichenized fungi, fungal diversity, chorology

**Introduction**

The lichenized fungi *Lecanora ramulicola* and *L. phaeostigma* have been generally considered rare in Poland. However, intensive field studies carried out by the first and third authors in large coniferous forests located in the Western Carpathians and the northern part of Poland revealed them to be quite common in this country. Moreover, detailed studies of herbarium materials referring to the *Lecanora symmicta*-group from the five biggest Polish collections show

that they had been collected in the past quite frequently but, for nomenclatural or taxonomic reasons, had never been published. In order to clarify this issue we initially focused only on these two species. During the study, however, we also found *L. aitema*, which has never been reported from Poland. In addition, we were interested in the possible occurrence of true *L. cadubriae* in this country, something that was unclear because of nomenclatural confusion; it was formerly synonymized in the Polish literature with both *L. ramulicola* and *L. phaeostigma*. Thus, here we elucidate the current status of *L. aitema*, *L. cadubriae*, *L. phaeostigma*, and *L. ramulicola* in Poland.

### Materials & methods

The studied material originated from the following Polish herbaria: GPN, KRA, KRAM, KRAP, and UGDA. The morphology and anatomy of the specimens were examined using standard microscopic techniques. Secondary substances were analyzed by TLC (in solvents A, B, and C) according to the methods of Orange et al. (2001). Unknown fatty acids observed in *Lecanora ramulicola* samples were compared with angardianic/roccellic acid (extracted from *Lepraria caerulescens* (Hue) Botnen & Øvstedal), jackinic/rangiformic and norjackinic/norrangiformic acids (extracted from *Cladonia rangiformis* Hoffm. and *Lepraria jackii* Tønsberg). The localities are listed and mapped in the modified ATPOL grid square system (Cieśliński & Fałtynowicz 1993, see also Kukwa et al. 2002). The following abbreviations are used in the citation of localities: NP – National Park; fd. – forest district; fs. – forest section; vill. – village; vall. – valley; sl. – slope.

### The species

#### *Lecanora aitema* (Ach.) Hepp

PLATE 1A

SPECIMENS EXAMINED — POLAND, Carpathians: [Gd-17] – Pasma Babiogórskie range, Krowiarki pass, alt. ca 1020 m, on horizontal surface of spruce snag, 22.06.1965, Nowak (KRAM-L-15917); [Gd-27] – Pasma Babiogórskie range, Polica range, Zubrzyca Górna vill., Syhlecc vall., alt. ca 900 m, on decaying stump, 23.06.1965, Nowak (KRAM-L-17168); [Ge-12] – Beskid Wyspowy Mts., Mogielica Mt., alt. ca 1170 m, on wood of decaying spruce stump, 06.08.1966, Nowak (KRAM-L-4630); [Ge-21] – Gorce Mts., N sl. of Czoło Mt., alt. 1200 m, on horizontal surface of *Picea abies* snag in well-lit place within upper mountain spruce forest, 30.07.1967, Glanc (KRAM-L-29696).

*Lecanora aitema* has not yet been reported from Poland, perhaps due to its unclear taxonomic status. Specimens of *L. aitema* were frequently considered to represent darker forms of *L. symmicta*, since these taxa differ essentially by apothecial pigmentation alone. The taxonomic status of *L. aitema* needs molecular resolution, but currently more and more lichenologists treat this taxon as a separate species (see below). A revision of some Polish collections of

'*L. symmicta*' shows that several of them have almost exclusively dark, blackish-grey, blackish-brown, convex to subglobose apothecia (PLATE 1A) and a slightly greenish thallus containing usnic acid and zeorin. The anatomical characters of the apothecia agree well with those described by Wirth (1995) for *L. symmicta* var. *aitema* and by Edwards et al. (2009) for *L. aitema*. We therefore decided to report *L. aitema* as new for Poland. Some Polish specimens originally labeled *L. symmicta* var. *aitema* are, in fact, *L. ramulicola*. It is interesting that we found only old specimens of *L. aitema* in the herbarium collections of *L. symmicta*; recent collections have not been made despite intensive field studies.

CHEMISTRY — Usnic acid, zeorin.

ECOLOGY AND DISTRIBUTION IN POLAND — The old collections show that *L. aitema* occurs on hard wood of conifers, mostly on horizontal surfaces of spruce stumps within upper mountain spruce forests in the Carpathians. This corresponds well with known ecological preferences of the species mentioned by Edwards et al. (2009). Accompanying species include: *Cladonia* spp., *Biatora pullata*, *Lecanora* sp., *Micarea denigrata*, *Parmeliopsis ambigua*, and *Xylographa parallela*.

WORLD DISTRIBUTION — The precise distribution of this species is not known since its taxonomic position at the species level has sometimes been questioned (see, e.g., Nimis 1993). Often, this taxon has been treated as *Lecanora symmicta* var. *aitema* (Ach.) Th. Fr., *L. symmicta* var. *saepincola* (Ach.) Nyl., or *L. aitema*. *Lecanora aitema* has been synonymized with *L. symmicta* (Ach.) Ach. and, under the latter name, it was mentioned in several national and regional checklists (Wirth 1995, Pišút et al. 1996, Ciurchea 1998, Hafellner & Türk 2001, Bielczyk 2003, Fałtynowicz 2003, Clerc 2004, Mayrhofer et al. 2005, Kossowska 2006). The name *L. symmicta* var. *aitema* has also been incorrectly used for *L. ramulicola*. For these reasons, a re-examination of *L. symmicta* s. l. collections is required to uncover the real global distribution of *L. aitema*.

Distributional data presented here include only reports of *L. aitema* or *L. symmicta* var. *aitema*, as these most probably refer to this taxon. As suggested by Nimis (1993), *L. aitema* appears to be widespread, at least in boreal and montane European regions. Indeed, it is reported from Greenland (Alstrup et al. 2009), the Czech Republic (Liška et al. 2008), Denmark (Søchting et al. 2007), Germany (Scholz 2000, Kanz et al. 2005, Dolnik & Neumann 2009), Great Britain (Smith et al. 2009), Ireland (Seaward 1994), Italy (Nimis 1993), the Netherlands (Aptroot et al. 2004), Fennoscandia (Santesson et al. 2004), and the European part of Russia (Hermansson et al. 1998). Esslinger (2009) has also reported *L. aitema* for North America.

### *Lecanora cadubriae* (A. Massal.) Hedl.

PLATE 1B

EXSICCATAE EXAMINED — Lichenes Alpinum 287: AUSTRIA, Steiermark, Schladminger Tauern, alt. ca 1350 m, an *Larix decidua*, 09.07.1973, Poelt (KRAM-L-25797); 318: Austria, Kärnten, Tauren, Kreuzeck-Gruppe, alt. ca 1850 m, an Stämmen von *Larix decidua* am Waldrand, 15.07.1978, Wirth & Hertel (KRAM-L-25827); 376: ITALY, Südtirol: Zillertaler Alpen, Riesenferner-Gruppe, alt. 1850 m, an der Stamm-Basis einzeln am Hang stehender *Larix decidua*, 18.10.1979, Hertel (KRAM-L-25888); Rabenhorst, Lichenes europaei 731 & Massal. Lich. Ital. exs. N. 332!: Italy, Riva, sulla corteccia dei

Larici in varie localita, 1864, Abbé Carestia (KRA-17702); Lichenes Slovakiae Exsiccati 35: SLOVAKIA, Liptovské Tatry, ad corticem Laricis deciduae in monte Klinovate, alt. ca 1300 m, 10.08.1963, Vězda (KRAM-L-25902).

In the Polish literature (Bielczyk 2003), '*Lecanora cadubriae*' was synonymized under the illegitimate name *Lecidea ramulicola* H. Magn. published in 1952 (see Printzen & May 2002). The name '*Lecanora phaeostigma*' was also erroneously used as a synonym of *L. cadubriae* (Fałtynowicz 2003, Kossowska 2006) and it is probable that some published Polish reports of *L. cadubriae* refer to *L. phaeostigma*. However, most of the reported Polish samples of '*L. cadubriae*' appear to represent morphological forms of *L. ramulicola* with less developed apothecia than usual and a thick, cracked thallus (see under *L. ramulicola*). The characteristic secondary substances for *L. cadubriae* have not been detected in any available Polish specimen originally filed under this name. Thus its real occurrence in Poland is questionable. *Lecanora cadubriae* has been collected many times close to Poland in the Slovak Tatra Mts. (Lisická 2005). Considering this and other reports from Central Europe (e.g., Kanz et al. 2005, Liška et al. 2008) the Polish Tatra Mts. is the most probable area to discover the species for the national lichen biota.

**CHEMISTRY** — P+ orange, K+ yellow turning to orange; TLC: norstictic acid (major), ±stictic and ±salazanic acids (accessory substances).

**WORLD DISTRIBUTION** — The general distribution of *Lecanora cadubriae* extends from the boreal zone (including North America and Greenland) to the montane areas of Europe (Nimis 1993, Thomson 1997, Alstrup et al. 2009, Smith et al. 2009). The species appears in numerous European checklists and catalogues, being sparsely recorded inter alia from: the British Isles (Smith et al. 2009), the Nordic (Søchting & Alstrup 2002, Santesson et al. 2004) and the Baltic countries (Randlane & Saag 1999, Jüriado et al. 2003), Central Europe (Scholz 2000, Hafellner & Türk 2001, Kanz et al. 2005, Lisická 2005, Liška et al. 2008), the Balkans (Mayrhofer et al. 2005, Knezevic & Mayrhofer 2009) and the alpine regions of Italy and Slovenia (Nimis 1993, Suppan & Mayrhofer 2002). Furthermore, outside Europe and North America, the species was reported from Syria (John et al. 2004).

It seems that the lichen is rare but rather widespread in the Northern Hemisphere. It should be taken into account, however, that the name '*Lecanora ramulicola*' has sometimes been treated erroneously as a synonym of *L. cadubriae* and that some records of *L. cadubriae* refer, in fact, to *L. ramulicola*.

### *Lecanora phaeostigma* (Körb.) Almb.

PLATE 1C

**SELECTED SPECIMENS EXAMINED** — (if not otherwise stated, on wood of *Picea abies* within upper mountain spruce forest). POLAND, Carpathians: [Gd-16] – Western Beskidy Mts., Babia Góra Massif, Babia Góra NP, fs. no. 21A, 49°35'04.6"N / 19°32'15.3"E, alt. 1160 m, 03.07.2009, Czarnota 6156 (GPN); [Gd-17] – Babia Góra Massif, Babia Góra NP, fs. no. 18a, SE sl. of Sokolica Mt., 49°35'12.3"N / 19°34'07.8"E, alt. 1230 m, 08.06.2009, Czarnota 6044 (GPN); *ibid.*, fs. no. 25, S sl. of Sokolica Mt., 49°35'02.3"N / 19°33'56.3"E, alt. 1265 m, 01.07.2009, Czarnota 6093 (GPN); [Gd-26] – Babia Góra Massif, Babia Góra NP, fs. no. 26h, S sl. of Kępa Mt., 49°34'07.7"N / 19°33'01.9"E, alt.



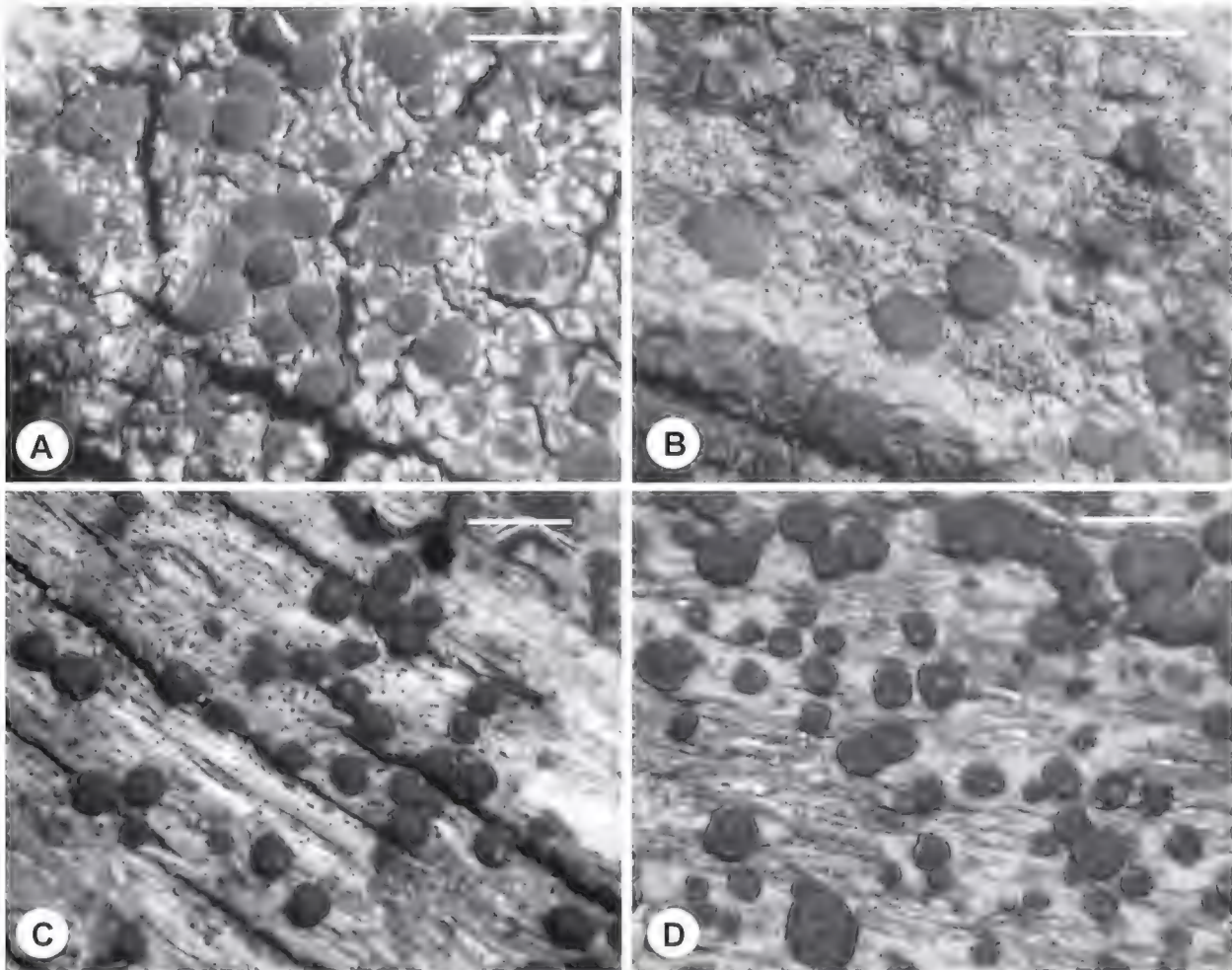


PLATE 1. Habits of discussed *Lecanora* species. A – *L. aitema* (KRAM-L-17168); B – *L. cadubriae* (KRA-17702); C – *L. phaeostigma* (GPN 6069); D – *L. ramulicola* (GPN 5771). Scale bars = 1 mm.

1365 m, 17.07.2009, Czarnota 6069 (GPN); [Gd-27] – Babia Góra Massif, Babia Góra NP, fs. no. 26h, S sl. of Kępa Mt., 49°34'07.8"N / 19°33'12.8"E, alt. 1290 m, 17.07.2009, Czarnota 6079 (GPN); [Ge-11] – Western Beskidy Mts., Gorce Mts., Gorce NP, W sl. of Gorce Kamienicki Mt. near Spaleniec stream, alt. 820 m, 27.03.2002, Czarnota 2754 (GPN); [Ge-20] – Gorce Mts., vall. of Lepietnica stream, W of Długie Młaki glade, alt. 1160 m, 12.09.1966, Glanc (GPN/5368; Ex KRAM-L-39562) and on bark of *Picea abies*, 12.09.1966, Glanc (KRAM-L-39186, as *Lecidea cadubriae*); *ibid.*, Bukowina, Dolina Robowa vall., alt. 715 m, on bark of *Picea abies* within *Abieti-Piceetum*, 21.07.1965, Glanc (KRAM-L-39187, as *Lecidea cadubriae*); [Ge-21] – Gorce Mts., Gorce NP, fs. no. 136a, Dolina Kamienicy vall. above Mały Borek region, 49°33.619'N / 20°09.098'E, alt. 980 m, within *Abieti-Piceetum*, 06.09.2008, Czarnota 5723 (GPN); *ibid.*, N sl. of Turbacz Mt. below the top, alt. 1280 m, 25.05.2001, Czarnota 5369 (GPN); *ibid.*, W sl. of Mostownica range, alt. 1220 m, 23.07.2007, Czarnota 5458b (GPN); *ibid.*, fs. no. 35d, E sl. of Czoło Mt., 49°33.160'N / 20°07.325'E, alt. 1200 m, 14.10.2008, Czarnota 5797 (GPN); [Ge-60] – Tatra Mts., High Tatra Mts., Tatra NP, NW sl. of Żabia Grań Mt. near the border Pl-Sk, alt. 1500 m, 09.07.2002, Czarnota 2874 (GPN, as *Lecidea hypopta*).

There are practically only historical records of *Lecanora phaeostigma* within Poland (Körber 1855, Stein 1879, Eitner 1901). These records accepted here are based only on the original and detailed descriptions included in Körber's and Stein's works. Stein included *Biatora phaeostigma* Körb., a basionym of



*L. phaeostigma*, as a synonym of *B. obscurella* (Sommerf.) Arnold. According to Zahlbruckner (1925), *B. obscurella* was based on *Lecidea pellucida* var. *obscurella* Sommerf. described in 1826 and raised to the species level as *Lecidea obscurella* (Sommerf.) Nyl. by Nylander in 1866. Hedlund (1892) mentioned this species as *Lecanora obscurella* (Sommerf.) Hedl. [nom. illegit. as a later homonym of *Lecanora obscurella* (J. Lahm.) Nyl. 1878], but surprisingly *Biatora obscurella* has recently been included in the synonymy of *Biatora tetramera* (De Not.) Coppins (Index Fungorum 2010), a species that is not congeneric with *Lecanora phaeostigma* in a current sense. In fact, the nomenclature of the taxa, not to mention the taxonomy, is highly complicated and warrants a separate study.

CHEMISTRY — 2-methylene-3-carboxy-18-hydroxynonadecanoic acid.

ECOLOGY AND DISTRIBUTION IN POLAND — *L. phaeostigma* grows frequently on hard wood of decorticate *Picea abies* trunks and, rarely, on the bark of usually dead spruces within upper montane spruce forest (*Plagiothecio-Piceetum*) and montane spruce-fir forest (*Abieti-Piceetum*) at altitudes between 820–1500 m. It prefers semi-shaded parts of trunks but tolerates well-lit localities within extensive insect damaged stands. Accompanying species usually include *Calicium abietinum*, *C. glaucellum*, *C. trabinellum*, *Fuscidea pusilla*, *Lecanora ramulicola*, *L. subintricata*, *Lecidea leprarioides*, *Parmeliopsis ambigua*, *Pycnora sorophora*, and *Strangospora moriformis*.

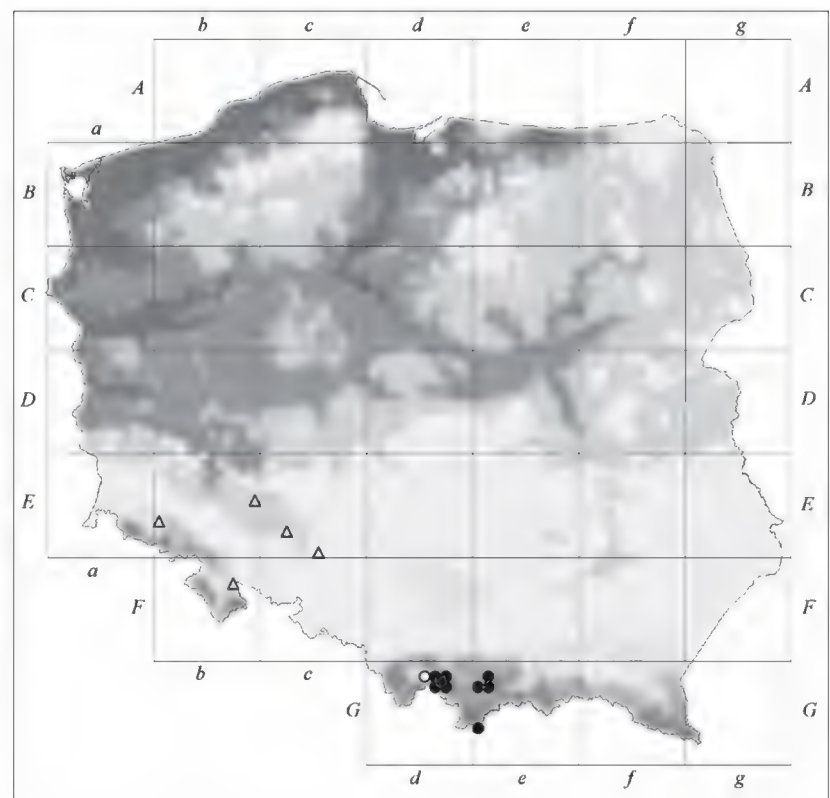


PLATE 2. Known distribution of *Lecanora phaeostigma* in Poland given in ATPOL grid square system (100×100 km): Δ – historical data; ○ – recent record; ● – new findings.

The name of *Lecanora phaeostigma* has been reported recently from Poland only twice – from a locality in the Western Beskidy Mts. (Nowak 1998) and from the Polish Eastern Carpathians in the Low Bieszczady Mts. (Kiszka & Kościelniak 1998). Unfortunately, there are neither detailed localities nor any ecological data given in the latter report. Moreover, the species was omitted in later lists compiled for that region (Kościelniak & Kiszka 2003, Kościelniak 2004), from which it has not been reported since 2004. The occurrence of *L. phaeostigma*, however, is possible in the Polish Bieszczady Mts. since it is known in the Ukrainian part of the Eastern Carpathians (Kondratyuk et al. 1998). PLATE 2 shows the known distribution of the species including the old reports cited above.

WORLD DISTRIBUTION — *L. phaeostigma* is regarded as a rare but rather widespread species in Europe. It was reported throughout the continent from Fennoscandia, Western Russia, and Estonia (Randlane & Saag 1999, Jüriado et al. 2003, Santesson et al. 2004, Urbanavichus et al. 2008), Romania and Bulgaria (Ciurchea 1998, Mayrhofer et al. 2005), Germany, Austria, and Switzerland (Scholz 2000, Hafellner & Türk 2001, Clerc 2004, Kanz et al. 2005, Pfefferkorn-Dellali & Türk 2005), and France and Italy (Clauzade & Roux 1985, Nimis 1993). Close to Poland it is found in the Czech Republic and Ukraine (Kondratyuk et al. 1996, Kondratyuk et al. 2003, Liška et al. 2008). Outside Europe, the species was recorded from Mongolia (Biazrov 2009).

***Lecanora ramulicola* (H. Magn.) Printzen & P.F. May**

PLATE 1D

≡ *Lecidea saepincola* var. *ramulicola* H. Magn.

≡ *Lecidea ramulicola* (H. Magn.) Hillm.

SELECTED SPECIMENS EXAMINED — (if not otherwise stated, on wood or bark of conifers: *Pinus sylvestris* or *Picea abies*). POLAND: [Ac-35] – Wybrzeże Słowińskie coast, Szklana Huta fd., fs. no. 164b, 2.5 km E of Lubiatowo vill., on dead twigs of *Betula pendula*, 18.09.2000, Kowalewska (UGDA-L-15484); *ibid.*, fs. no. 109f, 3 km NE of Lubiatowo vill., on *Betula pendula*, 9.10.2000, Kowalewska (UGDA-L-15488); [Ac-36] – Wybrzeże Słowińskie coast, Białogóra nature reserve, 27.07.1982, Fałtynowicz (UGDA-L-1461); Białogóra fd., fs. no. 25g, 1 km NW of Białogóra vill., on *Betula pendula*, 10.10.2000, Kowalewska (UGDA-L-15490); *ibid.* fs. no. 29b, 2 km NW of Białogóra vill., on *Betula pendula*, 25.08.2000, Kowalewska (UGDA-L-15481); *ibid.*, fs. no. 91g, 3 km W of Białogóra vill., on dead twigs of *Betula pendula*, 12.10.2000, Kowalewska (UGDA-L-15493); [Ac-38] – Wybrzeże Słowińskie coast, peat-bog Bielawskie Błoto, 30.09.1981, Fałtynowicz (UGDA-L-1401, as *L. symmicta*); [Ac-41] – Wybrzeże Słowińskie coast, Słowiński NP, Radek near Czołpino, 30.08.1988, Fałtynowicz (UGDA-L-3978, KRAM-L-22458, both as *L. symmicta*); [Ac-61] – Wysoczyzna Damnicka plateau, 1-2 km SE of Damnica vill., 20.11.1987, Fałtynowicz & Miądlukowska (UGDA-L-3752, as *L. symmicta*); [Bc-16] – Pojezierze Południowopomorskie lakeland, Bory Tucholskie forest, peat-bog near Drzędno Lake, 54°03'46"N / 18°00'34"E, 29.07.1983, Fałtynowicz (UGDA-L-1705, as *L. symmicta*); [Bc-25] – Bory Tucholskie forest, Grzybowski Młyn fd., fs. no. 365b, 2.5 km NE of Szludron village, 15.09.2002, Kowalewska (UGDA-L-15503); *ibid.*, 1 km NW of Szludron village, 19.06.2002, Kowalewska (UGDA-L-15500); *ibid.*, 0.5 km SW of Loryniec vill., 15.09.2002, Kowalewska (UGDA-L-15506); [Bc-26] – Bory Tucholskie forest, Lipa vill. at E shore of Wdzydze Lake, 53°59'24"N / 17°56'25"E, 13.09.2006,

Czarnota 5132 (GPN); [Bc-31] – Równina Charzykowska plain, „Niedźwiady” peat-bog nature reserve near Lipczynek vill., 26.07.1987, Fałtynowicz (KRAM-L-21769, UGDA-L-3108, both as *L. symmicta*); [Bc-65] – Bory Tucholskie forest, Biała fd., fs. no. 129, 16.08.2002, Czarnota 3077 (GPN); [Cg-64] – Nizina Północnopodlaska lowland, Równina Bielska plain, Białowieża Primeval forest, Hajnówka fd., fs. no. 572, 11.08.2002, Czarnota 3021 (GPN); [Fd-47] – Wyżyna Krakowsko-Częstochowska upland, Wyżyna Olkuską upland, Skalskie near Olkusz town, 06.04.1956, Nowak (KRAM-L-2556, as *Lecidea ramulosa*); [Fd-48] – Wyżyna Olkuską upland, Ojców vill., 01.05.1956, Nowak (KRAM-L-2446, as *Lecidea ramulicola*); [Fd-66] – Kotlina Oświęcimska depression, Rozkochów on Wisła River, 31.03.1960, Nowak (KRAM-L-6855, as *Lecidea ramulicola*); [Fd-93] – Western Carpathians, Western Beskidy Mts., Beskid Mały Mts., Przegibek pass, alt. ca 660 m, 23.08.1960, Nowak (KRAM-L-7583, as *Lecidea ramulicola*); [Fd-96] – Beskid Mały Mts., Łysa Góra Mt., alt. ca 510 m, close to the hiking track from Wadowice to Leskowiec, 19.04.1961, Nowak (KRAM-L-7065, as *Lecidea ramulicola*); [Fe-96] – Western Carpathians, Pogórze Rożnowskie foothills, Górowa vill., alt. 420 m, 11.09.1970, Kozik (KRAP, dupl. in KRA, as *L. cadubriae*); [Ff-03] – Kotlina Sandomierska basin, Puszcza Sandomierska forest, fs. no. 187, between Grębów and Stalowa Wola town, 07.09.1982, Kiszka (KRAP, as *Lecidea ramulicola*); [Ff-13] – Puszcza Sandomierska forest, Krawce fd., fs. no. 91, 28.09.1982, Kiszka (KRAP, as *Lecidea ramulicola*); [Fg-22] – Kotlina Sandomierska basin, Puszcza Solska forest, near Huta Różaniecka vill., 28.07.1984, Kiszka & Piórecki (KRAM-L, as *Lecidea ramulicola*); [Fg-23] – East Roztocze, Puszcza Solska forest, near Narol town, 20.07.1984, Kiszka (KRAM-L, as *Lecidea ramulicola*); [Gd-16] – Western Beskidy Mts., Beskid Żywiecki Mts., Babia Góra Massif, Babia Góra NP, Mała Babia Góra Mt., alt. 1460 m, 10.08.2001, Węgrzyn 379 (KRA, as *L. symmicta*); *ibid.*, Babia Góra NP, fs. no. 24a, 49°35'10.5"N / 19°30'52.3"E, alt. 1230 m, 02.07.2009, Czarnota 6130 (GPN); [Gd-17] – Babia Góra Massif, Babia Góra NP, Sokolica Mt., alt. 1300 m, 30.08.2000, Węgrzyn 192 (KRA, as *L. symmicta*); *ibid.*, fs. no. 25a, 49°35'01.0"N / 19°33'54.6"E, alt. 1265 m, 01.07.2009, Czarnota 6102 (GPN); *ibid.*, fs. no. 18b, NE sl. of Sokolica Mt., 49°35'19.5"N / 19°33'39.9"E, 10.06.2009, Czarnota 5999 (GPN); Beskid Żywiecki Mts., Polica Mt., alt. ca 1300 m, 8.07.1965, Nowak (KRAM-L-15145, as *L. symmicta*); [Gd-58] – Tatra Mts., West Tatra Mts., Wielkie Koryciska, alt. ca 1000 m, 19.06.1998, Bielczyk (KRAM-L-44435, as *L. symmicta*); [Gd-59] – Obniżenie Orawsko-Podhalańskie depression, Rów Podtatrzański depression, Dolina Lejowa vall., Polana Biały glade, 49°17'02"N / 19°50'48"E, alt. 900 m, 17.07.2004, Śliwa 3257 (KRAM-L-54579, as *L. symmicta*); [Ge-11] – Western Beskidy Mts., Gorce Mts., Gorce NP, N sl. of Kudłoń Mt., alt. 1130 m, 24.08.1967, Glanc (KRAM-L-48422, as *L. symmicta*); *ibid.*, vall. of Turbacz stream below Turbacz Mt., alt. 870 m, 04.11.1994, Czarnota 684 (GPN, as *L. cadubriae*); *ibid.*, W sl. of Kudłoń Mt. close to Pustak glade, 49°33'14"N / 20°10'13"E, alt. 1200 m, 11.07.2008, Czarnota 5371 (GPN); *ibid.*, vall. of Rosocha stream, alt. 780 m, 05.12.1994, Czarnota 672 (GPN, as *L. symmicta*); [Ge-20] – Gorce Mts., by hiking track from Turbacz Mt. to Nowy Targ-Kowaniec, 12.09.1964, Glanc (KRAM-L-28344, as *Lecidea helvola*); [Ge-21] – Gorce Mts., Gorce NP, Dolina Łopusznej vall., below Gabrowska glade, alt. 1240 m, 27.08.1968, Glanc (KRAM-L-40264, as *L. symmicta*); *ibid.*, fs. no. 136c, vall. of Kamienica stream below Jaworzyna glade, 49°33.415'N / 20°09.244'E, alt. 1075 m, 09.09.2008, Czarnota 5638 (GPN); *ibid.*, fs. no. 35a, W sl. of Mostownica Mt., 49°33.302'N / 20°07.318'E, alt. 990 m, 08.10.2008, Czarnota 5818 (GPN); *ibid.*, fs. no. 184b, W of Gabrowska glade, 49°32.624'N / 20°08.402'E, alt. 1240 m, 29.08.2008, Czarnota 5762 (GPN); [Ge-60] – High Tatra Mts., Tatra NP, N ridge of Żabia Grań Mt. on the border of Pl-Sk, alt. 1570 m, 09.07.2002, Czarnota 2921 (GPN);

ibid., ridge of Siedem Granatów Mt., 49°12'39.6"N / 20°05'25.0"E, alt. 1600 m, mountain stone pine and spruce forest, on *Pinus cembra*, 15.07.2006, Węgrzyn 3195 (KRA).

There are few literature records of *L. ramulicola* in Poland (Hillmann & Grummann 1957, Nowotarska 1976, Kozik 1977, Kiszka 1979, 1981a, b; Cieśliński et al. 1982 – all reported as *Lecidea ramulicola*; Printzen & May 2002, Kiszka 2008, Kubiak 2008 – reported as *Lecanora ramulicola*). Moreover, Nowak & Tobolewski (1975) mentioned that the species then occurred in the lowland and Carpathian foothills but did not include any detailed localities. Due to the nomenclatural and taxonomic confusion concerning this taxon between 1982 and 2002, there was a twenty-year-old gap in any information on *L. ramulicola* in Poland. Kiszka (1993) reflected that situation in Polish lichenology when he included *Lecidea ramulicola* as a synonym of *Lecanora cadubriae*. Since 1993, specimens of *L. ramulicola* have been usually cited in Polish literature exclusively (and erroneously) as *L. cadubriae* (e.g., Kiszka 1998, Kiszka & Grodzińska 2004). During our revision we found many specimens of *L. ramulicola* labeled as *L. cadubriae*. Some were originally labeled correctly as *Lecidea ramulicola* but later were annotated as *L. cadubriae* and thus never published. Although Printzen & May (2002) finally resolved the nomenclatural problems surrounding these species, distributional data of *Lecanora ramulicola* in Poland remained hidden under '*L. cadubriae*' (e.g., Bielczyk 2003), a species that probably has never been collected in Poland (see under *L. cadubriae*). Our revision showed also that *Lecanora ramulicola* was often misidentified as *Lecanora symmicta* (including *L. symmicta* var. *aitema*).

**TAXONOMICAL REMARKS** — Darker forms of *Lecanora ramulicola* resemble *L. aitema* because of the similar apothecial pigmentation. The biatorine apothecia of *L. ramulicola*, however, are usually distinctly marginate, ±glossy and often concave when immature (PLATE 1D). Moreover, the taxa are chemically distinct. According to Printzen & May (2002), *L. ramulicola* produces atranorin as a major compound together with one unknown substance. Some paler, mature, immarginate forms of *L. ramulicola* often resemble *L. symmicta*, but these species differ in the colour of thallus: distinctly ash-grey in *L. ramulicola* (because of the predominance of atranorin) and slightly yellowish green in *L. symmicta* (because of abundant usnic acid). In addition, the areoles of *L. ramulicola* are usually more coherent.

Printzen & May (2002) present an excellent description, and they discuss the affinities and differences between other taxa in the *Lecanora symmicta*-group (forming biatorine, mature apothecia and containing usnic acid as a major compound).

**CHEMISTRY** — Atranorin, ±usnic acid, 1 or rarely 2 unidentified fatty acids. Most of the analyzed specimens contain usnic acid, but often, only small

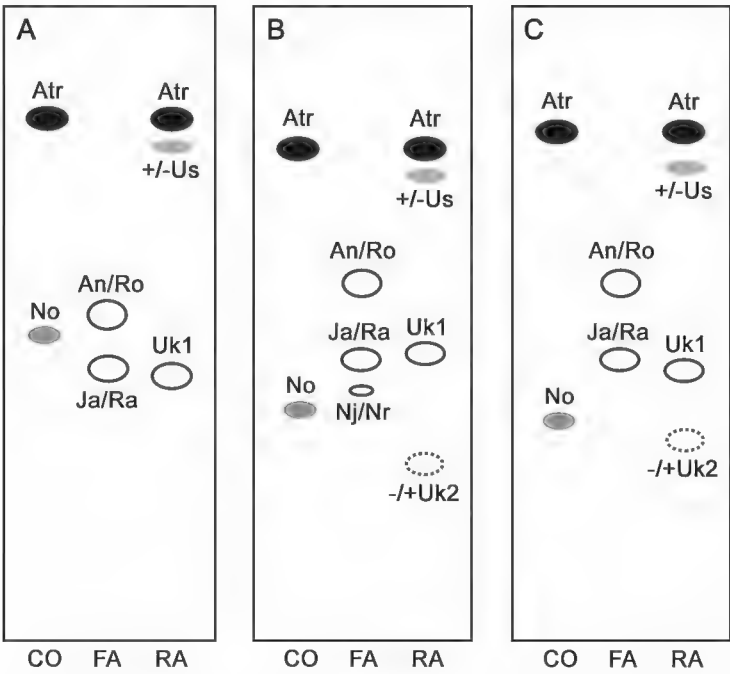


PLATE 3. Schematic diagram of chromatograms showing substances detected in *Lecanora ramulicola*, in solvent systems A, B, and C. CO – control; FA – selected fatty acids; RA – *L. ramulicola*. Compounds: An/Ro – angardianic/roccellic acid; Atr – atranorin; Ja/Ra – jackinic/rangiformic acid; Nj/Nr – norjackinic/norrangiformic acid; No – norstictic acid; Uk1 and Uk2 – unknown fatty acids; Us – usnic acid.

amounts of this substance were detected. Unknown fatty acids differ from substances mentioned in the Material and methods section (see PLATE 3). The first fatty acid (Rf classes A3-4, B4, and C4-5) is always produced whereas the second fatty acid (Rf classes A not detected, B2-3, C3) occurs sporadically. Only a dozen of the examined specimens contained trace amounts of this satellite substance.

ECOLOGY AND DISTRIBUTION IN POLAND — *Lecanora ramulicola* is widespread in Poland from the Baltic coast (Printzen & May 2002) to the Tatra Mts., and from close to sea level to the upper timberline at an altitude of 1500 m. Most of its known localities, however, lie to the south in the Western Carpathians. It usually grows there on the hard wood of branches and decorticate trunks of *Picea abies* within the upper montane spruce forest *Plagiothecio-Piceetum* or, more rarely, in the Carpathian beech forest of the lower montane belt. A few collections have also been made from the bark of conifers (e.g., *Picea abies*, *Pinus sylvestris*, *Abies alba*). It prefers well-lit places within dead stands destroyed by bark beetles and open localities at the edges of forest gaps. Large lowland pine forests are also favoured habitats. There, *L. ramulicola* grows both on bark and wood of *Pinus sylvestris* and frequently on the dead twigs of *Betula pendula*.

The known distribution of *L. ramulicola* in Poland is shown in PLATE 4. Localities from Upper Silesia mentioned by Kiszka (1993) for *Lecanora cadubriae*



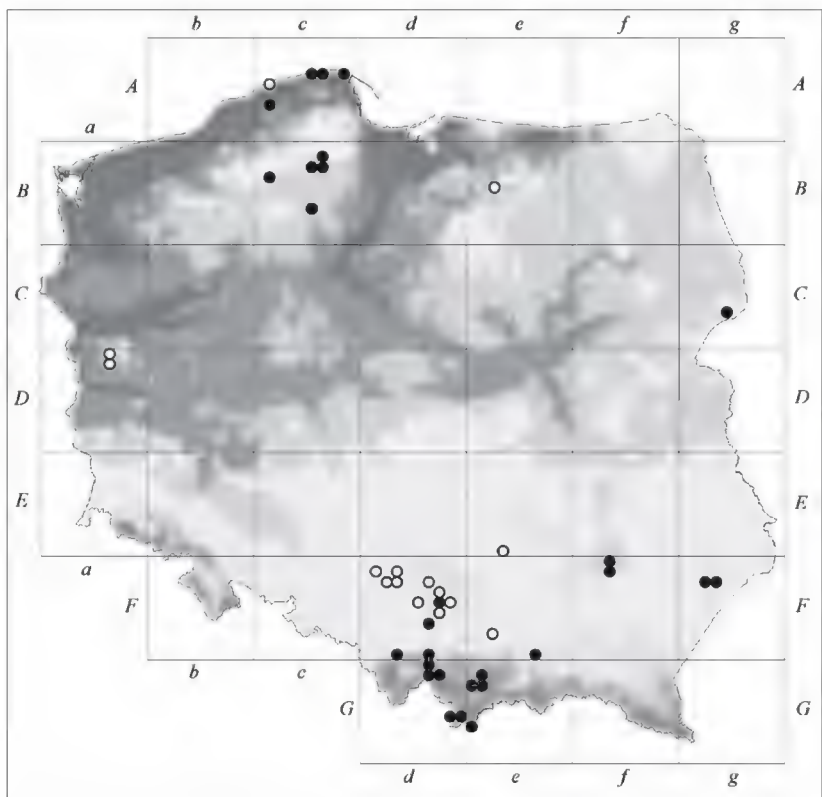


PLATE 4. Known distribution of *Lecanora ramulicola* in Poland given in ATPOL grid square system (100×100 km): ○ – previously reported localities; ● – new findings.

have been included here without taxonomic revision, since the specimens clearly refer to *Lecidea ramulicola*. Anyway, revision of those materials was impossible because the collection was unavailable in KRAP.

WORLD DISTRIBUTION — *L. ramulicola* was an overlooked lichen in the past and its general distribution is not yet well known. Printzen & May (2002) reported it from some central European countries (the Czech Republic, Poland, Germany) and North America (Canada and the U.S.A.). Later the species was reported from the Slovak part of the Tatra Mts. (Lisická 2005), the Iberian Peninsula (Pérez-Ortega & Printzen 2007), Western Russia (Kuznetsova et al. 2007) and some additional localities in Germany (Kanz et al. 2005, Dolnik & Neumann 2009).

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**The epitypification of *Ophiostoma minutum*,  
now *Ceratocystiopsis minuta***

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**Abstract** — Siemaszko's (1939) illustrations and figure legends for *Ophiostoma minutum* are designated herein as the lectotype for *Ceratocystiopsis minuta*, and a strain UAMH 11218 [= WIN(M) 1532, = R. Jankowiak 705] isolated from perithecia in galleries of *Ips typographus* in stems of *Picea abies*, from Biebrzanski National Park (Polish: Biebrzański Park Narodowy), Werklye Protection Range, grown and dried on wood chips, is then designated as the epitype and deposited in UAMH. This specimen will serve as a reference in future studies on species of *Ceratocystiopsis* that use modern morphological, chemotaxonomic, and molecular approaches. Morphological details are also presented for the epitype material.

**Key words** — nomenclature, ophiostomatoid fungi, species delimitation

**Introduction**

The genus *Ceratocystiopsis* H.P. Upadhyay & W.B. Kendr., based on *Ceratocystiopsis minuta* (= *Ophiostoma minutum*), was erected to accommodate ophiostomatoid fungi that have short perithecial necks and falcate ascospores (Upadhyay & Kendrick 1975). Siemaszko (1939) did not mention a conidial state for his fungus, and none of his material is extant. However Upadhyay (1981) indicated the anamorph of this species is a *Hyalorhinocleriella* H.P. Upadhyay & W.B. Kendr. Additional significant papers in the history of the erection of *Ceratocystiopsis* are those of Davidson (1942) and Mathiesen (1951).

Davidson (1942) took up Siemaszko's name as "*Ceratostomella* (*Ophiostoma*) *minutum* Siem." for several isolates he obtained from stained sapwood and grubs of *Monochamus titillator* (Fabr.) infesting a single dead pine near the District of Columbia, U.S.A. And while fertile perithecia were produced by his isolates, cultures he started from single ascospores were sterile; no perithecia were formed. He also recorded that this fungus produced a "cephalosporium-



like” conidial state, but that the perithecia it produced were smaller than those measured by Siemaszko.

Next Mathiesen (1951) provided an amended description for *O. minutum*, based on a number of the collections from both spruce and pine trees that had beetle galleries in them of one of four different bark beetles; perithecia were found regularly amongst the frass in the galleries. She noted some differences in that several of her measurements fell between those recorded by Siemaszko and Davidson. She also provided a detailed description of what she, too, called a “cephalosporium-like” conidial state, but her figures illustrate more complex fruiting structures than do those of Davidson.

Upadhyay’s (1981) treatment of the fungus, by then known as *Ceratocystiopsis minuta*, was based entirely on North American material, for he did not record a single off-shore specimen as having been examined. Yet while Davidson reported the perithecial necks were 45–90 µm in length, Upadhyay stated they were 45–150 µm long. He also assigned the conidial state to the genus *Hyalorhinocladella*, even though *Hyalorhinocladella minutibicolor*, the type species of that anamorphic genus, does not resemble the conidial state of *Ceratocystiopsis minuta* as figured by Mathiesen (1951). [The original spelling of the specific epithet “*minuta-bicolor*” has been corrected throughout in accordance with Articles 60.8 and 60.9 of the ICBN (McNeill et al. 2006).]

Historically, the genus *Ceratocystiopsis* was not accepted by Wingfield et al. (1988), Hausner et al. (1993), or Van Wyk & Wingfield (1993); indeed Hausner et al. (1993) formally reduced it to synonymy with *Ophiostoma*. Subsequently, Zipfel et al. (2006), who discussed the taxonomic placement of the falcate-ascospored, short-necked, ophiostomatoid fungi, re-instated the genus name *Ceratocystiopsis*. They placed eleven species in the genus, but addressed only briefly the taxonomic and phylogenetic inconsistencies that existed amongst the species, and it now appears likely that the specimen they selected to represent *Ceratocystiopsis minuta* in their study was an unfortunate choice; it was not explained. They also listed, with brief notes, eleven other species that might be linked in some way to accepted members of this genus.

Recently Plattner et al. (2009) reviewed the taxonomic and phylogenetic inconsistencies that surround strains and isolates of this species studied previously by various authors, and attempted to resolve them using a molecular-based approach. They made much progress, but the final result did not allow convincing conclusions to be drawn, although the extent of genetic diversity they uncovered within the complex showed clearly that several phylogenetic species have been combined under the name *Ceratocystiopsis minuta*. Unfortunately however, because neither herbarium material nor a viable culture exists that can be linked to Siemaszko’s (1939) original description of the basionym *Ophiostoma minutum*, they could make no further progress. Thus there is still no true nomenclatural type to serve as a reference for current workers.

Plattner et al. (2009) considered designating a neotype (new nomenclatural type) for *O. minutum* but were unable to do so because none of the cultures they considered to be appropriate candidate strains—these were from Poland-produced fully mature perithecia. However, using a modified culture technique and one of Plattner et al.'s designated candidate strains, R.J. 705 (UM 1532), we have obtained a very substantial number of mature perithecia on both wood chips and agar surfaces, but neotypification is not the appropriate course.

Along with his formal Latin description, Siemaszko provided photographs of two separate perithecia (possibly separate photographs of the same perithecium at different magnifications), a photograph of a bark sample with beetle galleries filled throughout with frass in which perithecia can be seen, and a line drawing of 22 ascospores showing their shape as enclosed within their mucilaginous sheaths. All these illustrations are referenced with his description and are certainly part of the material upon which the Latin description validating the name was based. Thus lectotypification based on these elements, followed by designation of an epitype, will serve to define this name.

## Materials and methods

Air-dried (20°C) wood chips were obtained from the face of the outer sapwood next to the inner bark of laboratory air-dried discs that had been cut from the stem of a healthy specimen of both *Picea glauca* (Moench) Voss and *Pinus sylvestris* L.; the chips, which ranged from 3.5–5 mm long, 1.5–2 mm wide, and up to 1 mm thick, were placed in a clean 600 ml beaker and flooded with enough of a solution containing 20 g malt extract, 1 g of yeast extract, and 0.02 g of thiamine hydrochloride per L distilled water to ensure the chips would be still fully covered when they became saturated. Next the beaker was placed in a sealed Nalgene Vacuum desiccator (Fisher Scientific, Fair Lawn, NJ), and the latter was then evacuated and allowed to stand overnight. The next day, after adding nutrient solution to ensure the chips were covered, the beaker was sealed with aluminum foil and autoclaved for one hour at 121°C. The stimulatory effect of thiamine on perithecial production has long been known (Barnett & Lilly 1947; Hawker 1957), and it has been used recently for this purpose with other ophiostomatoid fungi (van Wyk et al. 2004, 2006).

When cooled, in a sterile chamber the chips were aseptically placed flat on the medium surface of sterile Petri dishes; in the latter the medium had the same composition as the above nutrient solution, but with solidifying agar added at 20 g/L. Two separate inoculation series were undertaken; one used pine chips, the other spruce. Depending on the size of the chips, two to four were placed in each plate. Plates were then inoculated using 1 mm square blocks of mycelium cut aseptically from colony margins of stock plates of isolate RJ705. One block, mycelium face down, was placed immediately adjacent to each wood chip in a plate, and the plates were then incubated in the dark at 20°C for up to 60 days. In total, forty plates were inoculated: 18 with spruce chips (total 54) and 22 with pine chips (total 65).

Although *Ceratocystiopsis minuta* has been reported to occur on *Abies*, *Larix*, *Picea*, and *Pinus* spp. in association with a variety of bark beetle species, we used pine and spruce chips in our trials because spruce was the host recorded by Siemaszko (1939). Mathiesen (1951: 205), who recorded this species from both spruce and pine, observed that it occurred most commonly on pine, “meist auf Kiefer, weniger oft auf Fichte”.

Six agar plates containing only the amended malt extract agar plus thiamine hydrochloride were also inoculated centrally with single blocks of mycelium to serve as controls. Morphological structures were mounted in 85% lactic acid (Fisher Scientific, Fair Lawn, N.J.), and processed for observation according to Hausner et al. (2003). For photography we used Melzer's Reagent (Kohn & Korf 1975) to contrast the spore bodies with their surrounding sheaths.

## Results

As our purpose was to obtain appropriate material to serve as an epitype for *Ceratocystiopsis minuta*, only incidental cultural characteristics were recorded. Mycelium growth was slow, as Davidson (1942) and Mathiesen (1951) both noted with their isolates.

By day 10, the mycelium had grown onto the pine-wood chip surfaces, but less abundantly than onto the adjacent agar. On the agar surface it was white initially, but later became pale grey around the inoculation blocks; on wood, the mycelium remained white, and by this time small, pale grey, spherical bodies were present on both the wood and agar. Over the next 50 days many more spherical bodies formed in irregular patches on the mycelium on both the wood and agar surfaces, and a surprisingly large number of them matured into fertile perithecia.

Although initially the developing perithecial necks were darker than the perithecial bases, this slight “bicolored” condition disappeared as the perithecia continued to mature and was never as pronounced as that seen in perithecia of *Ceratocystiopsis minutibicolor* (R.W. Davidson) H.P. Upadhyay & W.B. Kendr. (Upadhyay & Kendrick 1975).

By day 20 there were abundant maturing/matured perithecia in localized patches on the agar and numerous more dispersed perithecia on the wood-chip surfaces. And by then the majority of the perithecia had become uniformly dark colored, and spore tendrils/droplets were seen at many neck apices (FIG. 1 A – E).

Over the next 40 days increasing numbers of mature perithecia formed, particularly on the wood chips, and at day 60 the plates were dried at 20 C in a drying oven, and stored for further study.

Although both Scots pine and white spruce chips were used in parallel inoculation trials, mature fertile perithecia formed only in the culture plates containing pine chips.

## Nomenclature

- Ceratocystiopsis minuta*** (Siemaszko) H.P. Upadhyay & W.B. Kendr.,  
Mycologia 67(4): 800. 1975 FIGS 1A–E, 2  
= *Ophiostoma minutum* Siemaszko, Plant Polonica 7: 23. 1939  
= *Ceratocystis minuta* (Siemaszko) J. Hunt, Lloydia 19: 49. 1956  
= *Ceratostomella minuta* (Siemaszko) R.W. Davidson, Mycologia 34: 655. 1942

## Lectotypification

The status of the genus *Ceratocystiopsis* H.P. Upadhyay & W.B. Kendr. is in doubt because no holotype specimen exists for the species upon which it was based, i.e. *Ceratocystiopsis minuta* (= *Ophiostoma minutum*); indeed none of Siemaszko's herbarium material is now extant. However, in accord with Article 9.2 of the ICBN (McNeill et al. 2006), Siemaszko's (1939) illustrations typify his description.

**LECTOTYPUS** (designated here): Siemaszko's *Planta Polonica* 7: plate III: 10, 11, 12; fig. 1B. 1939.

Siemaszko (1939) did not refer to any specific collection but merely the tree host species and beetle with which the fungus is associated in a specific geographical area. Plate III: 10 and 12 are photographs of two perithecia, 11 perithecia in beetle galleries, and Fig. 1B shows line drawings of ascospores.

## Epitypification

From the foregoing, the concept of *Ceratocystiopsis minuta* is now technically fixed, but Siemaszko's illustrations and description cannot possibly accommodate the needs of mycologists employing more modern morphological techniques or of those currently using chemotaxonomic and molecular methods to unravel relationships amongst various widespread populations of fungi such as *Ceratocystiopsis minuta*. Therefore we have chosen to designate an epitype.

**EPITYPUS** (designated here): UAMH 11218 (= a dried culture of WIN(M) 1532 = R. Jankowiak 705). ISOEPITYPE BPI 880579 (= R.Jankowiak(R.J. 705)/1532).

The strain was isolated from perithecia in galleries of *Ips typographus* (L.) in stems of *Picea abies* (L.) H. Karst., Biebrzanski National Park (Polish: Biebrzański Park Narodowy), Werklye Protection Range, Northeastern Poland, R. Jankowiak, date not given, and grown and dried by us on wood chips on agar.

## Description of the epitype

Perithecia on wood and agar form initially as pale grey, spherical bodies with a lighter coloured central area from which the neck develops. Young necks initially darker than the perithecial base, bicolored phase gradually disappears with maturity; the upper portion of the base darkens first, the



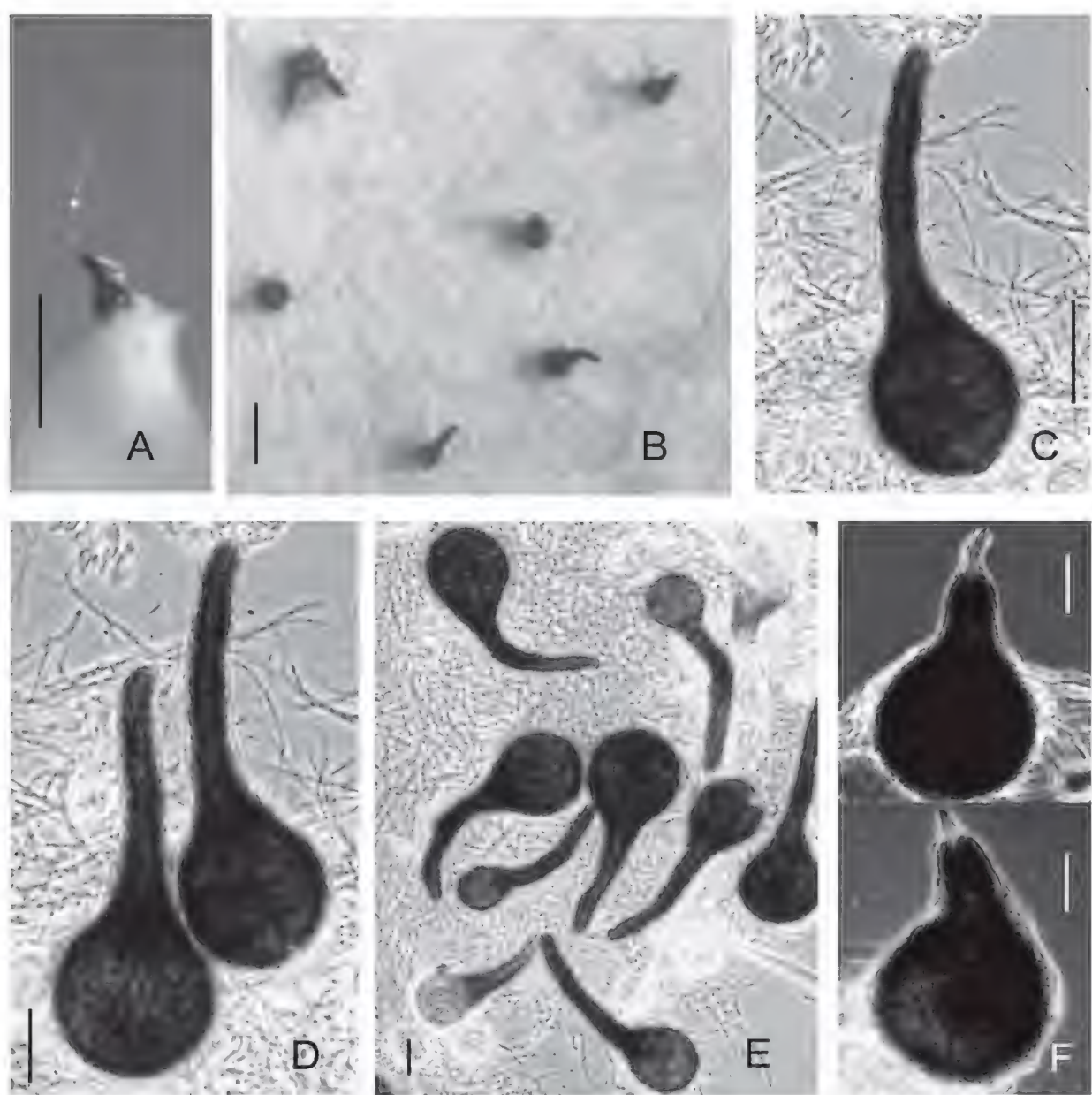


FIG. 1: A–E. *Ceratocystiopsis minuta* epitype UAMH 11218 [WIN(M) 1532]. A. A single perithecium, with an extruded spore tendril, on hyphal elements. B. Mature perithecia on the mycelium surface. C–E. Maturing and mature perithecia. Note the color variations in the young perithecia in E, and the extruded ascospores in D. F. *Ceratocystiopsis minuta* CBS 116795 [WIN(M) 1511]. Perithecia produced by this isolate in culture on wood; show the significant morphological differences apparent between the appearance of these perithecia and those produced by UAMH 11218. Scale bars: A and B = 250 µm; C–F = 30 µm.

darkening spreading downwards over time. Mature perithecial bases globose to obpyriform, surface smooth to slightly irregular, very rarely with short brown hairs, dark-brown to black in color; base width 37.5–87.5 (sd =  $59.41 \pm 13.17$ ) µm, base height 37.5–97.5 (sd =  $58.7 \pm 14.58$ ) µm. Perithecial necks with smooth to irregular surfaces, 20–45 (sd =  $28.89 \pm 6.8$ ) µm wide at the base, tapering to 7.5–17.5 (sd =  $13.8 \pm 4.98$ ) µm and narrowest at the apex; 70–175 (sd =  $113.4 \pm 17.98$ ) µm long, ostiolar hyphae not included. Ostiolar hyphae up to 12 µm long; 1.5 µm wide at the base and tapering to a slightly blunted point; hyaline



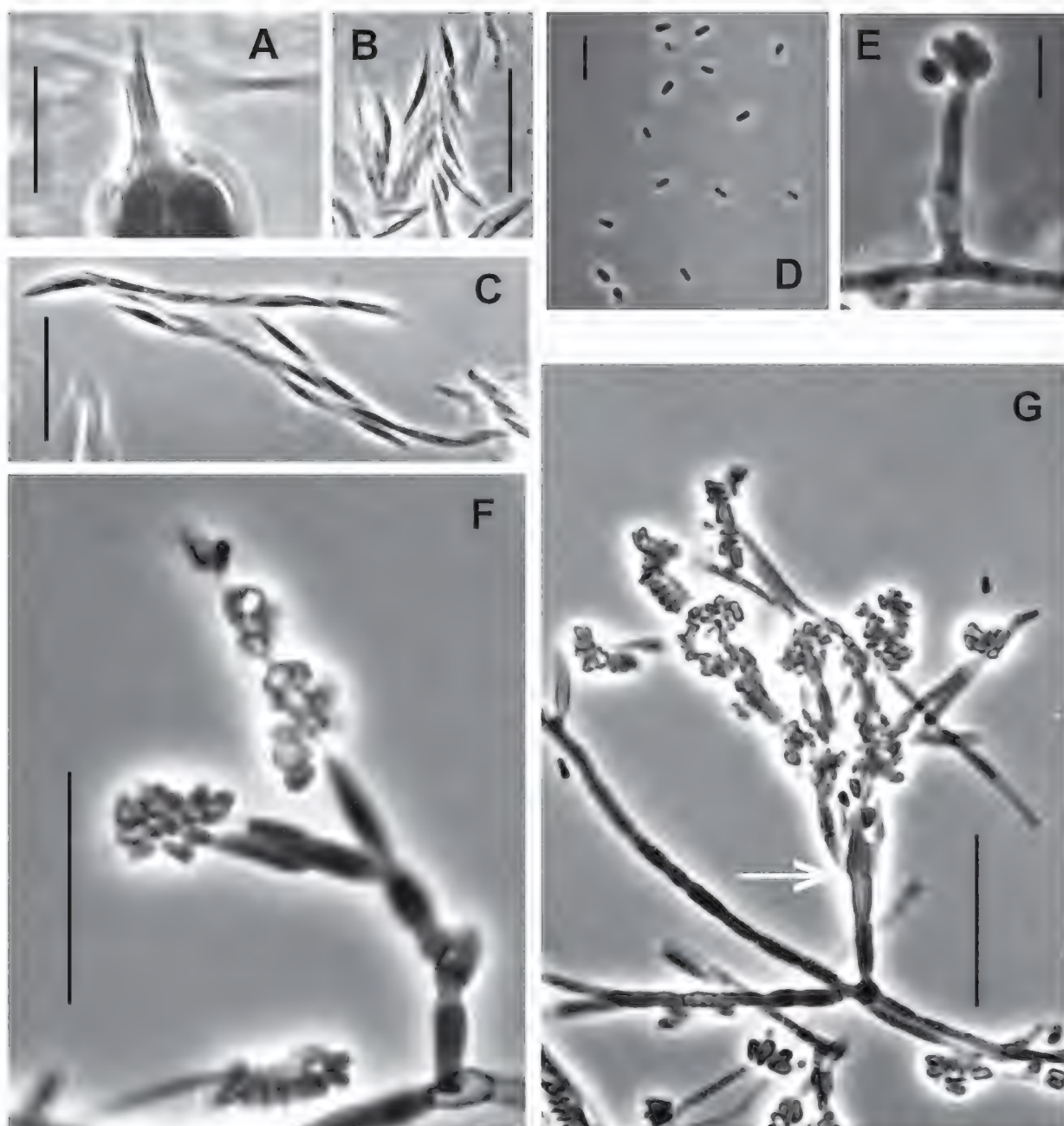


FIG. 2: *Ceratocystis minuta* epitype UAMH 11218. A. Neck apex of a mature perithecium with ostiolar hyphae. B and C. Ascospores stained with Melzer's reagent. D. Conidia. E. Simple conidiophore with adhering conidia. F. Branched conidiophore. G. Complex (macronematous) branching conidiophore; whorled branches origin is denoted by arrow. Scale bars: A, B and C = 20  $\mu\text{m}$ ; D and E = 10  $\mu\text{m}$ ; F and G = 30  $\mu\text{m}$ .

and convergent. Asci deliquesce rapidly and are seen rarely in mounting media; the free ascospores are usually extruded in long tendrils from the ostiole, but sometimes they collapse into mucilaginous droplets). Ascospores with sheath appear falcate, 10–13 (sd =  $11.4 \pm 1.18$ )  $\mu\text{m}$  long and widest, 1–2 (sd =  $1.38 \pm 0.48$ )  $\mu\text{m}$ , at their mid-point, tapering sharply to their tips. Ascospores without the sheath rarely seen.

Mycelium on agar white initially, becoming pale grey centrally and in infrequent patches with maturity. Aerial mycelium sparse, producing two types

of conidiophores. First type short, simple, randomly produced, with droplets containing numerous conidia at apices. Second type arise as short hyphal elements that continue to grow producing verticillate to irregular lateral and secondary branches. Conidiogenous cells appearing holoblastic, terminal, and percurrent. Conidia produced in slime drops, one-celled; oblong to slightly tapered with rounded ends, oval; small, 2–4 ( $sd = 3.34 \pm 0.59$ )  $\mu\text{m}$  long and 1–2 ( $sd = 1.38 \pm 0.48$ )  $\mu\text{m}$  wide hyaline and smooth.

We do not accept the synonymization of *Ceratocystis dolominuta* H.D. Griffin (Griffin 1968) with *Ceratocystiopsis minuta* (Upadhyay 1981). We concur with Griffin and Olchowecki & Reid (1974) that its consistently shorter ascospores separate it from *Ceratocystiopsis minuta*.

## Discussion

Ideally a designated epitype would be based on an isolate from the Białowiecki Park Narodowy (Białowiecki National Park) area of eastern Poland, as this is where Siemaszko (1939) made his collections. Therefore, Plattner et al. (2009) sequenced three strains isolated from hosts in that area by T. Kirisits in June, 2002; specifically CBS 116795, 116796 and 116963. Of these, only strain 116795, closely allied to 116963 in their tree, fruited in culture, but it did not fit well with either Siemaszko's protologue for *Ophiostoma minutum*, or the latter as amended by Mathiesen (1951); 116796 did not fruit either and grouped with four other Polish strains and one Japanese strain in a different clade of Plattner et al.'s tree (2009). None of these four latter Polish isolates were from Białowieża, but three were from northeastern Poland, between 150 and 210 miles north northeast of that locality. Plattner et al. (2009: 884) noted that one of these "... , R.J. 705, which produced what appeared to be mature perithecia but no ascospores, showed an imperfect state almost identical to that described by Mathiesen (1951) and Davidson (1942)" would be a candidate for neotypification if it had produced ascospores. It is this strain we used for epitypification.

In Europe, *Ceratocystiopsis minuta* is associated commonly with a wide range of bark beetles on more than one host tree species (Kirisits 2004). In some cases there is a common association between the bark beetle and this fungus, e.g. *Ips typographus* and *Ceratocystiopsis minuta* on *P. abies* in certain areas of France (Viri & Lieutier 2004), but in other geographical areas the association is rare. For example, during a study of the fungi associated with *Tomicus piniperda* (L.) attacking *P. sylvestris* at eight locations in Poland, Jankowiak (2006) found it at only one of the locations sampled, and then only at very low levels. Clearly, *Ceratocystiopsis minuta* is beetle-vectored, although with more than one species (Kirisits 2004).

Plattner et al. (2009) concluded that the name *Ceratocystiopsis minuta* referred to several phylogenetic species, and that different species misidentified as *Ceratocystiopsis minuta* might be present in different geographical locations. Our results indicate that populations of these putative different phylogenetic species may coexist within fairly restricted geographical areas; based on combined morphological and molecular criteria, this appears to be the case in northeastern Poland.

The molecular analysis in Plattner et al.'s tree (2009; FIG. 1) suggests that two distinct populations of *Ceratocystiopsis minuta* exist in eastern Poland. Isolate R.J. 705, which we have designated as epitype, is representative of one of these populations, while CBS 116795 belongs to the other. Their molecular distinctness is confirmed by the morphological differences that we observed between these two isolates (TABLE 1, FIGS. 1 A–F).

TABLE 1. Comparison of perithecial and ascospore measurements for *Ceratocystiopsis minuta* strains R.J. 705 and CBS 116795.

ISOLATE	R.J. 705	CBS 116795
PERITHECIUM (µm)		
Base width	37.5–87.5; sd = 59.41 ± 13.7	52.5–100; sd = 78.44 ± 13.03
Base height	37.5–97.5; sd = 58.7 ± 14.58	57.5–100; sd = 77.39 ± 12.58
Neck length	70–175; sd = 113.4 ± 17.98	30.0–75; sd = 49.8 ± 12.65
Neck base width	20–45; sd = 28.89 ± 6.8	20–35(–37.5); sd = 26.81 ± 5.0
Neck tip width	7.5–17.5; sd = 13.8 ± 4.98	(15–)17.5–25; sd = 20.56 ± 2.34
Ostiolar hyphae	up to 12. 4 in length	up to 12.5 in length
ASCOSPORES (µm)		
Length	10–13; sd = 11.4 ± 1.18	9–15; sd = 10.86 ± 1.34
Width	1–2; sd = 1.19 ± 0.29	1–2; sd = 1.14 ± 0.26
CONIDIOPHORE TYPE	macronematous & micronematous	micronematous

Although not precisely the same, another unusual situation is evident in reports of *Ceratocystiopsis minuta* from Japan. Plattner et al. (2009) noted that Japanese strains of this fungus from two different tree species, *Picea jezoensis* (Siebold & Zucc.) Carrière (Yamaoka et al. 1997) and *Larix kaempferi* (Lamb.) Carrière (Yamaoka et al. 1998) shared a common phylogenetic ancestor, although they were placed in two distinct monophyletic groups (clades). These two isolates, JCM 9367 (YCC-139) from *P. jezoensis* and JCM 9816 (YCC-294) from *L. kaempferi*, are similarly morphologically distinct from each other in both their perithecial appearance and the nature of their anamorphs (Yamaoka et al. 1997, Figs. 1–5; Yamaoka et al. 1998, Figs. 2–5), as isolate R.J. 705 is from CBS 116795. Also, JCM 9367 (YCC-139), is placed in the same clade as R.J. 705

in Plattner et al.(2009, Fig.1), and resembles the latter isolate in morphological features, i.e. perithecial neck shape and conidiophore complexity.

Upadhyay & Kendrick (1975) erected the genus *Hyalorhinocladiella*, based on *Hyalorhinocladiella minutibicolor*, to accommodate the anamorph of *Ceratocystiopsis minutibicolor*. While their description and photographs do represent accurately the conidial state of *Ceratocystiopsis minutibicolor* as described by Davidson (1966), their concept of *Hyalorhinocladiella* does not fit the anamorph of *Ceratocystiopsis minuta* as defined herein.

Benade et al. (1996) emended the description of conidiogenesis in *Ceratocystiopsis minutibicolor* to percurrent and sympodial extensions of the conidiogenous cell, but did not indicate whether the conidiophores could be more complex than the simple hyphal elements described and/or figured by Davidson (1966) or Upadhyay & Kendrick (1975).

Our photographs of isolate R.J. 705, plus those of isolate YC-139 (Yamaoka et al. 1997) and the drawings of Mathiesen (1951) of *Ceratocystiopsis minuta* all show that this species, as epitypified herein, most commonly produces quite complex conidiophores; the simple *Hyalorhinocladiella*-like structures are less abundant.

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## MYCOTAXON

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***Tylopilus oradivensis* sp. nov.: a newly described member of the *Tylopilus balloui* complex from Costa Rica**TODD W. OSMUNDSON<sup>1, 2</sup> & ROY E. HALLING<sup>1</sup>

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**Abstract** — Surveys of macrofungi associated with neotropical *Quercus* forests in Costa Rica resulted in the discovery of a *Tylopilus* species similar to *T. balloui* in appearance but exhibiting differences in typical basidiome size, basidiome coloration, basidiospore size and shape, typical pleurocystidial shape, and DNA sequence characters. Molecular data suggest that *T. oradivensis* and *T. balloui* share a relatively recent common ancestor and lend support to the hypothesis of a biogeographic connection between oak forests of the eastern United States and Central America. The orthographic correction of the epithet *ballouii* to *balloui* is hereby made in accordance with the International Code of Botanical Nomenclature.

**Key words** — *Boletaceae*, *Boletineae*, boletes, ectomycorrhizal fungi, *Rubinoboletus*

**Introduction**

Since the mid-1990s, a concerted effort to document macrofungal diversity in Costa Rican montane *Quercus* forests has yielded descriptions of previously unknown boletes and field data valuable for assessing biogeographic patterns in ectomycorrhizal fungi (Amtoft et al. 2002; Halling 1999, 2001; Halling & Mata 2004; Halling et al. 2004; Halling & Mueller 2002, 2003, 2005; Mueller & Halling 1995; Osmundson et al. 2007). Subsequent field and laboratory studies of Australian and Southeast Asian boletes have revealed a number of taxa morphologically similar to the distinctive North American species *Tylopilus balloui* (Peck) Singer but differing in several morphological as well as molecular

characters — these data suggest that the name *T. balloui* as commonly ascribed to field and herbarium collections represents a species complex rather than a single widespread species (Halling et al. 2008), an observation consistent with those of Watling (Watling 2001; Watling & Gregory 1988). A closer examination of Costa Rican collections from Cartago and San José provinces led to the discovery of a taxon morphologically and genetically distinct from *T. balloui*, which we here describe as *Tylopilus oradivensis*.

### Materials and methods

Macromorphological descriptions were made from fresh basidiomes. Alphanumerical color designations correspond to Kornerup & Wanscher (1967), and are noted as combinations of plate, column, and row numbers (e.g., 8A5). Micromorphological examinations were performed using air-dried tissue from field collections or herbarium specimens. Preparation of hand sections for observation of micromorphological characters and use of descriptive terms follow Largent et al. (1977). Basidiospore measurements are presented as  $(k-)m-n(-p)$ , where  $k$  is the smallest observed value,  $p$  is the largest observed value,  $m$  is the 5th percentile value, and  $n$  is the 95th percentile value (Tulloss & Lindgren 2005). Length-to-width ratio ( $Q$ ) of the basidiospores is presented in the same manner. Mean length ( $L_m$ ), width ( $W_m$ ) and length-to-width ratio ( $Q_m$ ) are presented with their standard deviations (sd). Voucher specimens were deposited in the herbaria of the New York Botanical Garden (NY) or the Field Museum (F), with duplicate collections deposited in the herbarium of the University of Costa Rica (USJ) (acronyms from Thiers 2010).

A comparison of nuclear ribosomal large subunit (nrLSU) DNA sequences between Costa Rican collections and United States *T. balloui* accessions was made using data from Halling et al. (2008). Sequences were downloaded from GenBank (accession numbers EU430731 (CR), EU430732 (CR), EU430734 (USA), and EU430737 (USA)) and aligned using MAFFT (Katoh et al. 2005). Alignments were trimmed at the 5' and 3' ends (<10 bp per end) in order to eliminate terminal gaps using MacClade 4.08 (Maddison & Maddison 2001), and the alignment was examined manually.

### Taxonomic description

***Tylopilus oradivensis*** Osmundson & Halling, sp. nov.

FIGS. 1, 2

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*Aspectu similis* *Tylopilus balloui*, sed pileo e stipite aurantiaco-rubro magi, basidiomatibus minori, basidiosporis longioribus e subfusiformibus ad fusiformibus vel formibus capsicorum jalapensis revocans.

**HOLOTYPE:** R.E. Halling 7562 (NY 136973), 28 May 1996, approximately 4.5 km east of km 31 of Interamerican Highway, Palo Verde, El Guarco, Cartago, Costa Rica, elev. 1600 m.

**ETYMOLOGY:** The epithet *oradivensis* (ora = coast; dives = rich, -ensis = suffix indicating origin or place) refers to Costa Rica as the origin of the type collection.

**ICONES:** Halling & Mueller (2005: 72, as *Tylopilus ballouii*).

**MACROCHARACTERS** — **PILEUS** 3–5.5(–6) cm broad, convex to plano-convex, dry, matted subtomentose or tomentose, garnet brown (9D8) to orange red (8B7), capsicum red, tomato red or English red (8C7 to 8A-E8); margin inrolled, sterile. **CONTEXT** white to cream (4A3), unchanging. **ODOR** not distinctive. **TASTE** mild. **TUBES** adnate, up to 6 mm deep, yellowish white (4A2) or paler. **PORES** concolorous with tubes, staining light yellowish brown or pale brown when bruised. **STIPE** 5–7 cm long, 0.8–1 cm thick, nearly equal or tapering toward base, glabrous or finely to heavily pruinose, concolorous with pileus or sometimes paler near a pale salmon (6A-B5-4) or white with orange (6A-B8-7) patches, becoming sordid pale; base white or pale cream-colored, developing pale yellow or pale brown stains from handling. Obscure coarse reticulum at apex (as viewed under hand lens) observed in one collection.

**MICROCHARACTERS** — **BASIDIOSPORES**  $(7.6\text{--})8.2\text{--}12(\text{--}13.6) \times (2.6\text{--})3\text{--}4(\text{--}4.4)$   $\mu\text{m}$  ( $L_m = 9.74$ ,  $sd = 1.13$ ;  $W_m = 3.47$ ,  $sd = 0.36$ ;  $Q = (2.1\text{--})2.23\text{--}3.5(\text{--}4.57)$ ;  $Q_m = 2.83$ ,  $sd = 0.39$ ), subfusiform, fusiform or jalapeño pepper-shaped, longitudinal axis often sigmoid; thin-walled, hyaline or pale yellow in 3% KOH; uniguttulate, but droplet often irregular and with the appearance of being the product of fusion of two or more individual droplets, inamyloid, pale cinnamon brown in deposit. **BASIDIA** clavate, 4-sterigmate, hyaline. **PLEUROCYSTIDIA** of the pseudocystidia type; thin-walled, narrow, subclavate or ventricose-rostrate,  $40\text{--}76(\text{--}90) \times 8\text{--}15(\text{--}18)$   $\mu\text{m}$ ; one collection (REH 7562) with larger ( $92\text{--}110 \times 20\text{--}38$   $\mu\text{m}$ ), ventricose-rostrate cystidia present; contents granular and golden brown in 3% KOH, exhibiting a positive reaction with acidic fuchsin. **CHEILOCYSTIDIA** clavate, strangulated, or isomorphic with pleurocystidia, with contents similar to pleurocystidia. **PILEIPELLIS** a loose trichoderm or mixtocutis, hyphae 4–7  $\mu\text{m}$  broad, thin-walled, hyaline; end cells cylindrical, occasionally apically constricted, with yellow-orange uniform contents that are oily in appearance, exhibiting positive acid fuchsin reaction. **PILEAL TRAMA** of interwoven cylindrical, thin-walled, smooth hyphae. **HYMENOPHORAL TRAMA** boletoid, bilateral; hyphae 5.6–9.6  $\mu\text{m}$  broad; lateral stratum elements parallel or slightly divergent; mediostratum hyaline to pale orange-brown, darker than lateral stratum especially when young. **STIPITPELLIS** a broken hymeniform layer or with closely spaced fascicles of end cells; end cells subclavate or clavate; hyaline; larger ventricose-rostrate caulocystidia occasionally present in some samples. **CAULOCYSTIDIA**  $48\text{--}66 \times 8\text{--}14(\text{--}17)$   $\mu\text{m}$ , clavate or ventricose-rostrate, thin-walled, hyaline or with orange-brown contents in KOH. **CLAMP CONNECTIONS** absent.

**ECOLOGY, RANGE, AND DISTRIBUTION** — Scattered to gregarious, associated with *Quercus* in montane forests (observed at elevations of 1600–1850 m) of the Talamanca Mountains, Costa Rica.



FIG. 1. Basidiomata of *Tylopilus oradivensis*, R.E. Halling 7562 (NY).

ADDITIONAL SPECIMENS EXAMINED — COSTA RICA. CARTAGO: El Guarco. PALO VERDE, approximately 4.5 km east of km 31 of Interamerican Highway, elev. 1600 m, 24 June 2000, R.E. Halling (*Halling* 7920) (NY); 1 June 2001, R.E. Halling (*Halling* 8087) (NY); ESTRELLA, approximately 5 km east of km 31 of Interamerican Highway, elev. 1685 m, 21 July 1993, R.E. Halling (*Halling* 7044) (NY); 15 November 1993, R.E. Halling (*Halling* 7170) (NY); 31 May 1994, R.E. Halling (*Halling* 7214) (NY); 14 June 1996, R.E. Halling (*Halling* 7681) (NY); 1999, G.M. Mueller (*Mueller* 4853) (F); 13 June 2001, R.E. Halling (*Halling* 8187) (NY). SAN JOSÉ: CASAMATA, approximately 1 km west of Interamerican Highway at Casamata on road to San Cristobal Norte, elev. 1850 m, 18 October 1994, R.E. Halling (*Halling* 7380) (NY).

COMMENTS — Both macro- and micromorphologically, *T. oradivensis* bears a close resemblance to *T. balloui*, described from the northeastern United States. The latter is here orthographically corrected from the originally published epithet *ballouii* in accordance with article 60.11 of the International Code of Botanical Nomenclature (2006 Vienna Code), as well as from the incorrect epithet *balonii* as published by Saccardo (1925). The correct orthography appears in Heinemann & Rammeloo (1983), although the original spelling “ballouii” remains in nearly universal use. *Tylopilus oradivensis* and *T. balloui* are both characterized by having a pale hymenophore, pileus and stipe coloration in shades of orange, short (compared to many other boletes) basidiospores that are hyaline or pale yellow in 3% KOH, and the presence of an oily orange pigment in the hyphae of the pileipellis. However, the newly described taxon differs from *T. balloui* in



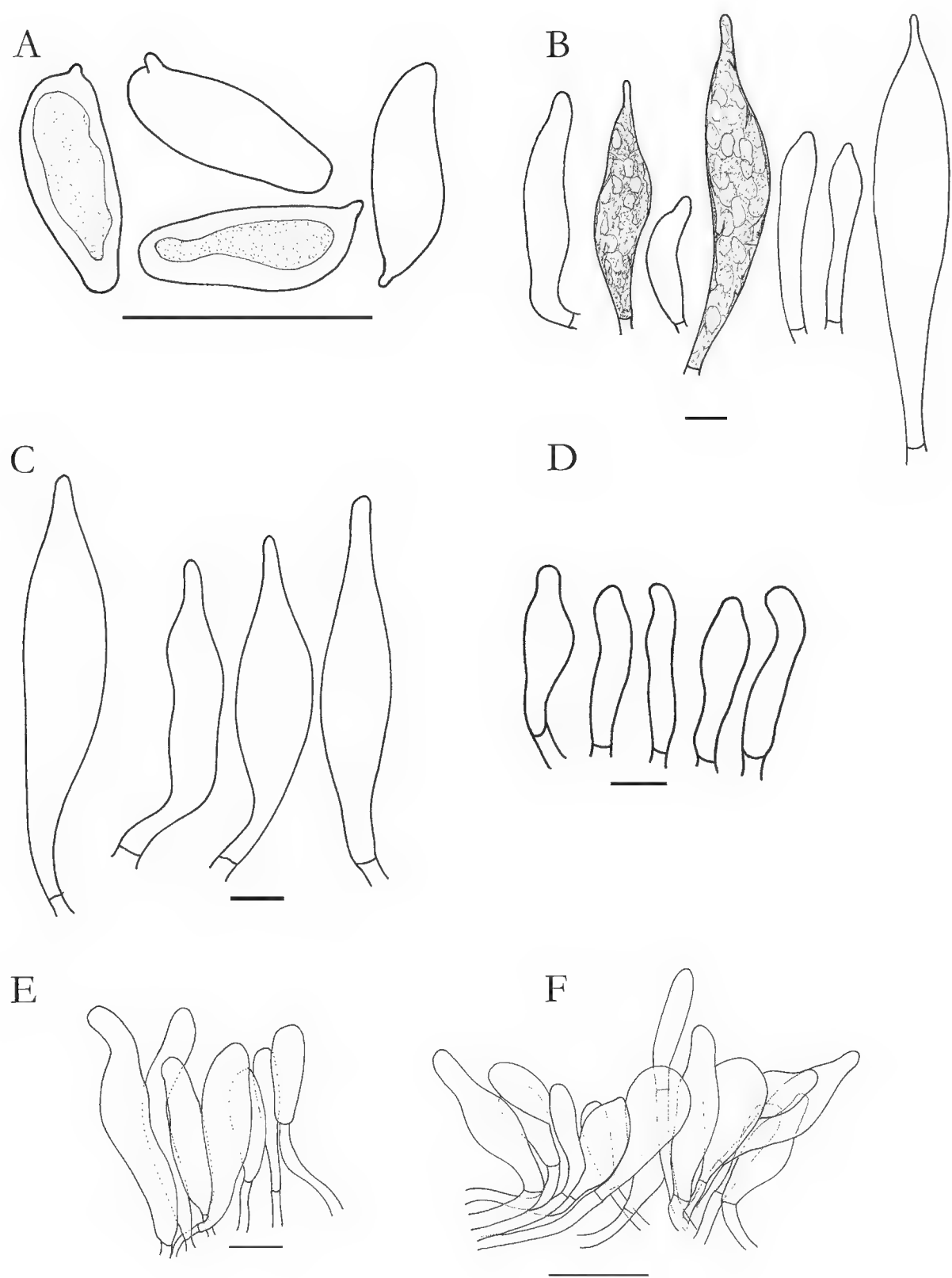


FIG. 2. Micromorphological features of *Tylopilus oradivensis*.  
a. Basidiospores (R.E. Halling 7562, HOLOTYPE). b. Pleurocystidia (R.E. Halling 7562, HOLOTYPE).  
c. Pleurocystidia (G.M. Mueller 4853). d. Cheilocystidia (R.E. Halling 7562, HOLOTYPE).  
e. Stipitipellis (R.E. Halling 7562, HOLOTYPE). f. Stipitipellis (R.E. Halling 7170).  
Scale bars: 10  $\mu$ m.

several respects. The original description of *T. balloui* by Peck (1912, as *Boletus ballouii*) describes a species with a bright orange to orange-brown pileus 5–12 cm broad, stipe 2.5–12 cm long and 0.7–1.5 cm thick, and basidiospores  $8\text{--}10 \times 4\text{--}5 \mu\text{m}$  that are clearly ellipsoid in Peck's original illustration. The Costa Rican material has smaller basidiomes (pileus  $<5.5\text{--}6$  cm), a reddish orange to nearly red (rather than bright orange) pileus (see comparison at <http://www.nybg.org/bsci/res/hall/oradivensis.html>), larger and differently shaped basidiospores, and pleurocystidia that are generally slightly narrower and longer than those in *T. balloui*. The end cells in the pileipellis are often tapered in *T. balloui*, but not in *T. oradivensis*. Although both Peck's description and the description of *T. balloui* by Singer (1947) note a range of spore dimensions that overlaps with that of *T. oradivensis* (dimensions recorded as  $7.5\text{--}11 \times 3.7\text{--}4.8 \mu\text{m}$  by Singer), both Peck's illustration and Singer's description strongly indicate that shorter, ellipsoid basidiospores are the normal condition. Wolfe's (1981) type study reports a range of basidiospore dimensions of  $5\text{--}10.5 \times 4\text{--}5 \mu\text{m}$ , with a mean of  $6.5 \times 4 \mu\text{m}$ . We have examined a large number of collections of *T. balloui* from the eastern, midwestern, and southern United States — including a collection made by Ballou (*W.H. Ballou s.n.*, accession 45249, NY), who provided a number of the collections of this taxon originally sent to Peck — and we have noted pileus dimensions of 4–9 cm, stipe  $3\text{--}10 \times 1\text{--}2.5$  cm, and broadly ellipsoid basidiospores  $6.4\text{--}8.4\text{--}8.8 \times (3.2\text{--})3.6\text{--}4.4 \mu\text{m}$ . Where larger basidiospores occur, they are rare and differently shaped (more ellipsoid) than those of *T. oradivensis*.

Nuclear ribosomal large subunit sequences differed between *T. oradivensis* and United States *T. balloui* in 23 of the 1421 nucleotide positions compared (98.4% sequence identity), with sequence differences comprised of 12 transitions, 2 transversions and 9 indel positions (five indels of 1, 1, 1, 2, and 4 bp). Sequences were invariable between the two collections within each species. While within-taxon sampling is too sparse to allow drawing conclusions regarding the genetic limits of the two taxa, these limited data nonetheless support the conclusion drawn from morphological analyses; i.e., that the two taxa are closely related yet distinct.

Both *Tylopilus oradivensis* and *T. balloui* would be placed in *Rubinoboletus* by some authors (Heinemann & Rammeloo 1983). However, *T. balloui* bears little morphological resemblance to the type species of *Rubinoboletus* (*R. rubinus* (W.G. Sm.) Pilát & Dermek, basionym *Boletus rubinus* W.G. Sm.) outside of having short, elliptical basidiospores. Singer (1947), in transferring *T. balloui* from *Gyrodon* (*G. balloui* (Peck) Snell) to *Tylopilus*, wrote: "The short spores do, in fact, occur in almost all groups of boletes and are not characteristic for *Gyrodon* alone." The same argument can — and we believe should — be applied

to placement of *T. balloui* within *Rubinoboletus*. *Tylopilus balloui* and its close relatives (including *T. oradivensis*) are indeed somewhat enigmatic among *Tylopilus* species; however, they are united with other species in the genus by several morphological characters including a pale hymenophore and hymenial pseudocystidia with dark yellow to brown pigmented contents (as observed in KOH mounts). Phylogenetic placement within a core *Tylopilus* clade is indicated by the nucLSU analysis of Binder & Hibbett (2006), and in our analyses using multiple loci (Osmundson et al. manuscript in prep.).

Consistent with the high sequence similarity observed between *T. oradivensis* and *T. balloui*, a previous phylogenetic analysis of nrDNA sequences for a broad geographic sample of *T. balloui* s.l. (Halling et al. 2008) indicated that the two species share a relatively recent common ancestor and lends support to the hypothesis of a biogeographic connection between oak forests of the eastern United States and Central America. As was hypothesized in the case of the species pair *T. chromapes* (*Leccinum chromapes*) and *T. cartagoensis* (*L. cartagoense*) (Wolfe & Bougher 1993), the close similarity between *T. oradivensis* and *T. balloui* would be consistent with a history of postglacial southward migration followed by morphological (and molecular) differentiation.

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## MYCOTAXON

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***Strelitziana africana* newly recorded from China**

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**Abstract** — We document the first report of *Strelitziana africana* from China. This fungus was isolated from stems of *Dioscorea cirrhosa* and *Sabia parviflora* collected from Nanning, Guangxi Province. *Strelitziana africana* can produce flyspeck signs on inoculated apple fruit and is distinguished from the other known species in the genus by morphological characters and phylogenetic analysis based on ITS sequences.

**Key words** — biodiversity, *Dioscoreaceae*, *Sabiaceae*, reservoir hosts, sooty blotch

**Introduction**

Sooty blotch and flyspeck (SBFS), a disease complex comprising more than 60 putative species of fungi, colonizes the waxy cuticle of many plants in humid production regions worldwide, inciting cosmetic damage that causes significant economic losses (Batzner et al. 2005). The common name “flyspeck” refers to species in the SBFS complex whose morphology on fruit surfaces consists of clusters of black dots lacking a mycelial matrix. Colby (1920) reported that flyspeck was caused by *Leptothyrium pomi* (Mont. & Fr.) Sacc. Von Arx (1959) synonymized 14 species, including *Leptothyrium pomi*, under *Schizothyrium pomi* (Mont. & Fr.) Arx, the currently accepted name for this taxon. Arzanlou & Crous (2006) reported *Strelitziana africana* Arzanlou & Crous from leaf speckle symptoms of *Strelitzia* in South Africa. The genus *Strelitziana* Arzanlou & Crous was named after the host genus from which it was collected and shown to be a member of the *Chaetothyriales*. We have identified two isolates that

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are described as the first record of *Strelitziana africana* from China based on morphological comparison and their phylogenetic relationships, as shown by analysis of sequence data of the internal transcribed spacer (ITS) of the rRNA repeat (Harrington & Rizzo 1999).

The species *Dioscorea cirrhosa* and *Sabia parviflora* are economically important medicinal plants in China because their stems and leaves are used as ingredients in Chinese traditional medicine. Recently, during a survey of alternate host plants for flyspeck fungi in China, these medicinal plants were found to be reservoir hosts of flyspeck fungi, including *Strelitziana africana*.

### Materials and methods

**ISOLATES.** Individual sclerotium-like bodies (Batzer et al. 2005), growing in clusters on culms, were transferred to slants containing potato-dextrose agar (200 g peeled potato, 20 g dextrose, 10 g agar in 1 L water; PDA) and cultured at  $22 \pm 1^\circ\text{C}$  in darkness (Sun et al. 2003). Colony descriptions were made after 1 month of growth on oatmeal agar (3%; 30 g oatmeal, 10 g agar in 1 L water; OA) plates at  $22 \pm 1^\circ\text{C}$  in darkness. After 1-month-old axenic cultures were transferred to new OA plates, a sterile cover slip was partially inserted into the agar adjacent to the colony and angled away from the colony at approximately 60 degrees to the agar surface in order to enable the fungus to grow onto the cover slip. Measurements of fungal structures were conducted based on isolates growing on cover slips.

**DNA SEQUENCING.** Template DNA was extracted from the fungal mycelium according to the method of Barnes et al. (2001), and primer pairs used for amplification and sequencing of the ITS region were ITS1-F (Gardes & Bruns 1993) and ITS4 (White et al. 1990). Amplification was completed with the following cycling parameters: initial denaturation at  $94^\circ\text{C}$  for 3 min followed by 35 cycles of denaturation at  $94^\circ\text{C}$  for 30 s, annealing at  $52^\circ\text{C}$  for 30 s, and extension at  $72^\circ\text{C}$  for 30 s and a final extension of  $72^\circ\text{C}$  for 10 min. The PCR products were sequenced by Organism Technology Co., Ltd., Shanghai, China.

The ITS nucleotide sequences generated in this study were added to sequences downloaded from GenBank (TABLE 1) that had high similarity according to a BLAST search (National Center for Biotechnology Information's nucleotide blast program). Preliminary alignments were performed using CLUSTAL-X (Thompson et al. 1997). The alignments were imported into BioEdit v. 5.0.9.1 (Hall 1999) and manually adjusted. Phylogenetic analysis of aligned DNA sequences was performed with PAUP v. 4.0b10 for 32-bit Microsoft Windows (Swofford 2001). Heuristic searches were performed with 1000 random sequence additions. Clade stability was evaluated by 1000 bootstrap replications. Other measures for parsimony, including tree length, consistency index, retention index, and rescaled consistency index (CI, RI and RC, respectively), were also calculated. *Curvularia affinis* was used as the outgroup taxon.

**KOCH'S POSTULATES.** After the two isolates in this paper were grown on OA for 1 month, a piece of the colony was picked up into an 1.5 ml Eppendorf tube and blended with 1.0 ml sterile deionized water for 1 minute. This suspension of mycelial fragments

TABLE 1. Sequences used in the phylogenetic analysis

SPECIES	GENBANK	REFERENCE
GX01	GQ850385	This paper
GX02	GQ850386	This paper
<i>Capronia acutiseta</i>	AF050241	Untereiner & Naveau 1999
<i>Capronia pulcherrima</i>	AF050256	Untereiner & Naveau 1999
<i>Cladophialophora devriesii</i>	AB091212	Abliz et al. 2003
<i>Cladophialophora</i> sp.	EU137326	De Hoog et al. 2007
<i>Curvularia affinis</i>	EF187909	Di Maiuta & Schwarzentruher 2007
<i>Mycosphaerella vietnamensis</i>	DQ632675	Burgess et al. 2007
	DQ632678	Burgess et al. 2007
<i>Pseudocercospora syzygiicola</i>	AF309600	Crous et al. 2000
<i>Pseudocercospora</i> sp.	DQ303082	Crous et al. 2006
<i>Rhinocladiella anceps</i>	AY163559	De Hoog et al. 2003
	DQ826740	De Hoog et al. 2003
<i>Rhinocladiella atrovirens</i>	AY163558	De Hoog et al. 2003
<i>Rhinocladiella basitona</i>	AY163561	De Hoog et al. 2003
<i>Rhinocladiella similis</i>	AY857529	Prenafeta-Boldu et al. 2006
<i>Strelitziana africana</i>	DQ885895	Arzanlou & Crous 2006
<i>Strelitziana australiensis</i>	GQ303295	Cheewangkoon et al. 2009

and conidia was used within 2 hours. Three mature, non-wounded apples were chosen for each isolate, surface-sterilized with 75% ethanol and allowed to dry completely, then swabbed with suspension of one isolate per apple. Two control apples were surface-sterilized and swabbed with sterile deionized water only. All the inoculated apples were incubated in a moist chamber at 22 ± 1°C.

Results

DNA phylogeny

A multiple alignment of the rDNA-ITS was generated with 18 sequences obtained from GenBank plus the sequences of isolates GX01 and GX02 (GX01 = GQ850385, GX02 = GQ850386). From a MP tree with a length of 750 bp (CI = 0.7080, RI = 0.7830, RC = 0.5512), two major clades were resolved (FIG. 1). One clade, with 100% bootstrap support, contained three species in *Mycosphaerella* and *Pseudocercospora*. The other major clade had a bootstrap value of 93%. The *Strelitziana* species grouped in a well-supported subclade (100%). Our isolates and a *Strelitziana africana* isolate from *Strelitzia* that was identified by Arzanlou & Crous (2006) fell within a single clade with 100% bootstrap support, indicating that they might represent the same species.

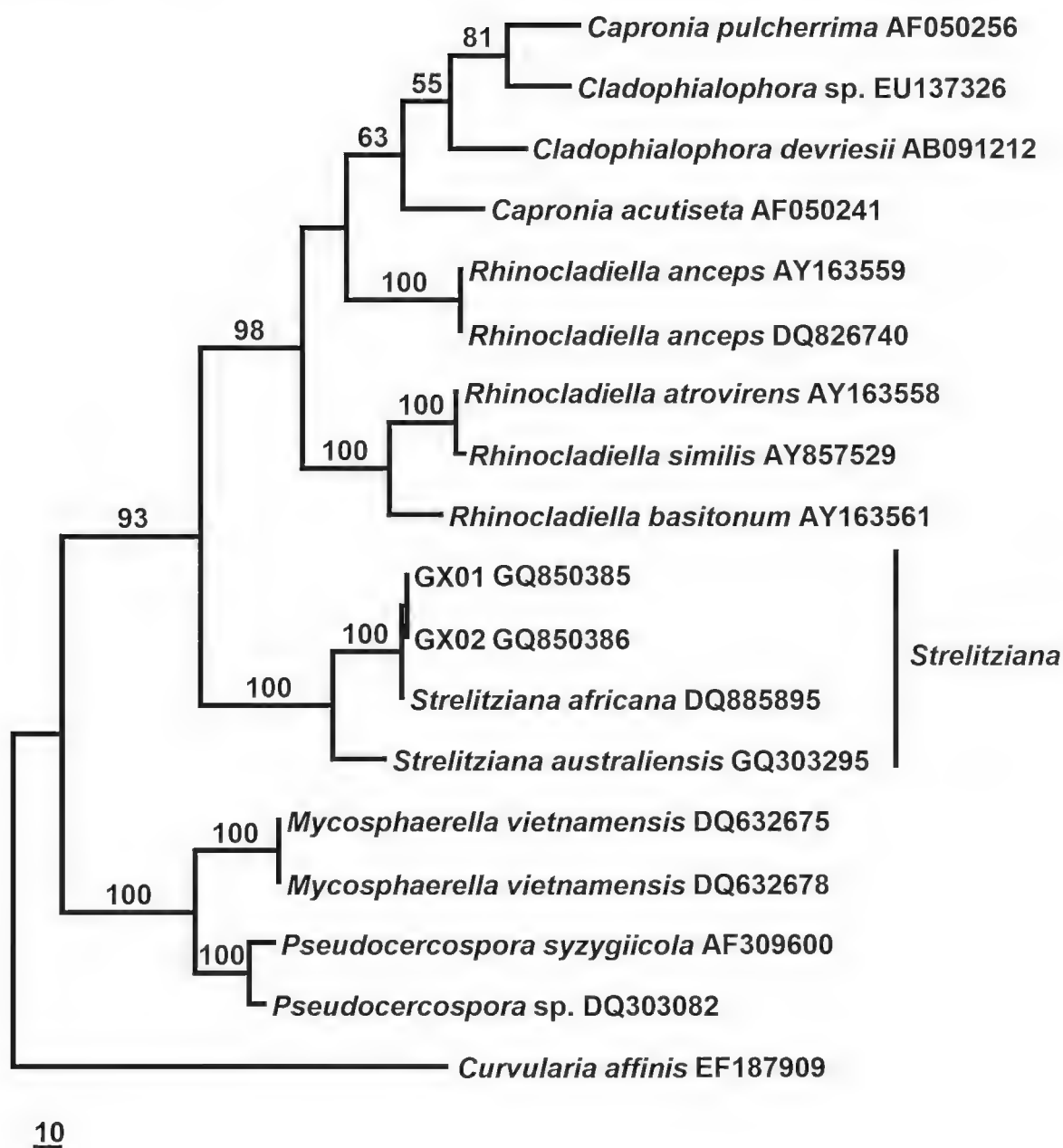


FIG. 1 The parsimony tree (TL = 750, CI = 0.7080, RI = 0.7830, RC = 0.5512) derived from a heuristic search option in PAUP v. 4.0b10 with 1000 randomizations of sequence input orders and 1000 bootstrap replications using the data set of ITS1, 5.8S and ITS2. Bootstrap values higher than 50% are indicated above the tree branches. The tree was rooted to *Curvularia affinis*.

### Taxonomy

DESCRIPTION: Mycelium superficial, consisting of smooth, septate, branched hyphae, 2–3 µm wide. Conidiophores erect, solitary, arising from aerial and submerged mycelium, subcylindrical, straight to geniculous-sinuous, pale brown, concolorous with hyphae, smooth, 0–4-septate, 3–20(–50) × 1.5–4.5 µm. Conidiogenous cells terminal, integrated, rejuvenating percurrently, proliferating apically via several short, conspicuous denticles, conidiogenesis rhexolytic. Conidia pale brown, smooth, long obclavate, widest in middle of



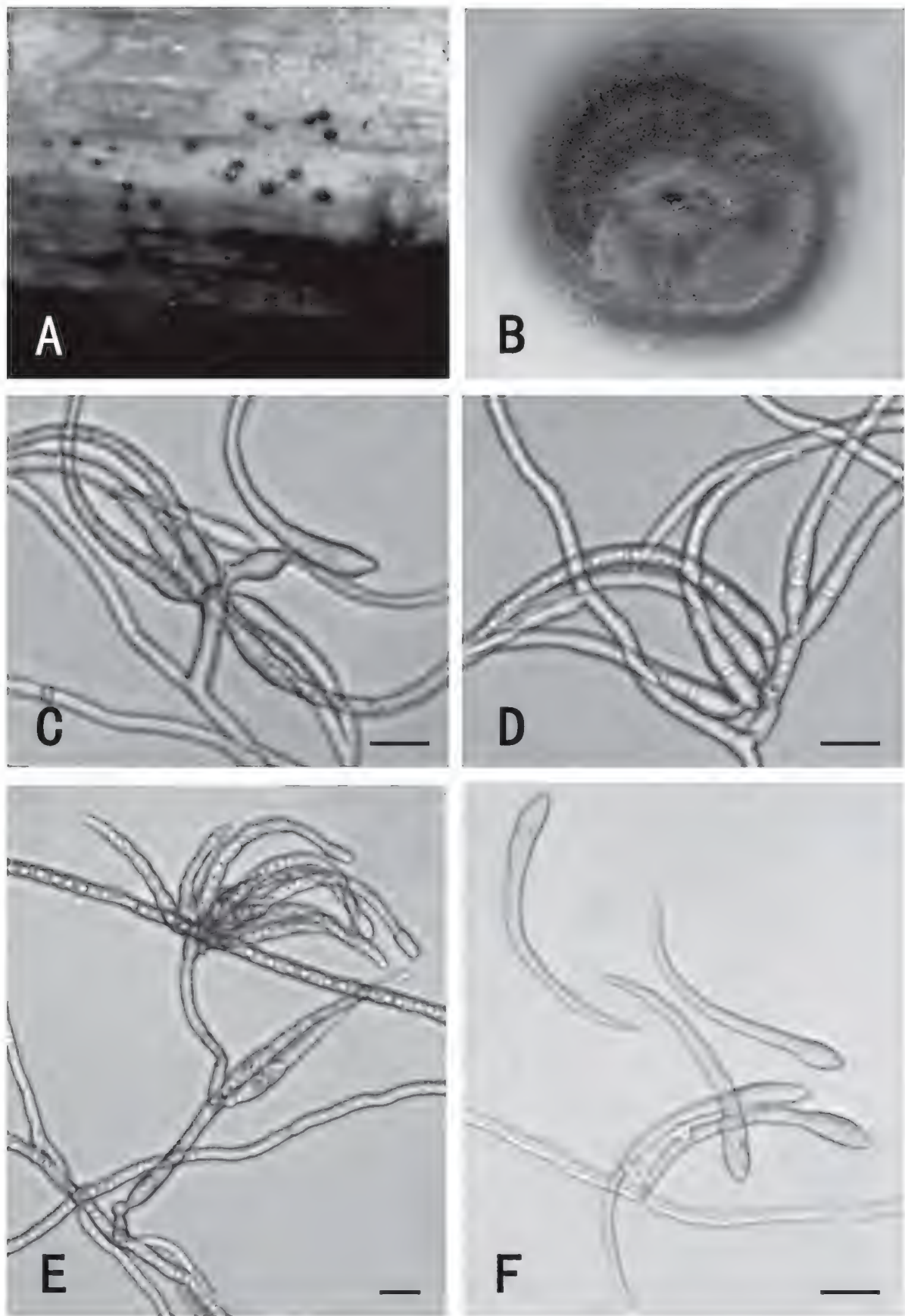


FIG. 2 *Strelitziana africana* GX01. A. Signs on *Dioscorea cirrhosa*. B. Colony on oatmeal agar after 30 days. C–D. Conidia, conidiogenous cells and hyphae. E. Conidiogenous cell giving rise to conidia, and microcyclic conidia. F. Conidia. Bars (C–F) = 10 μm.

basal cell, tapering to a subobtusely rounded apex and obconically subtruncate base with minute marginal frill,  $1\ \mu\text{m}$  wide,  $(12\text{--})35\text{--}70(100) \times 2.5\text{--}5\ \mu\text{m}$ ,  $3\text{--}10(21)$ -septate, microcyclic conidiation observed in culture (FIG. 2).

SPECIMENS EXAMINED: On *Dioscorea cirrhosa* Lour. (*Dioscoreaceae*), China: Guangxi, Nanning, Guangxi Medicinal Botanical Garden,  $22^{\circ}51'N$   $108^{\circ}19'E$ , alt. 72 m, 23 Sep. 2008, J.L. Zhuang & H.L. Yang, HMUABO (the Fungal Herbarium of Northwest A&F University) 822516 (with dried culture), culture GX01. On *Sabia parviflora* Wall. (*Sabiaceae*), China: Guangxi, Nanning, Guangxi Medicinal Botanical Garden,  $22^{\circ}51'N$   $108^{\circ}19'E$ , alt. 76 m, 23 Sep. 2008, J.L. Zhuang & H.L. Yang, HMUABO 822517 (with dried culture), culture GX02.

CULTURAL CHARACTERISTICS: The colony diameter after 1 month on PDA at  $22 \pm 1^{\circ}\text{C}$  reached 28 mm with even margins and smooth, felty aerial hyphae; colony centers were purplish gray and outer zones pale white. On OA the colony was flat, spreading, with sparse aerial mycelium, reaching 33 mm diam after 1 month at  $22 \pm 1^{\circ}\text{C}$ , surface olivaceous, colonies fertile.

HOST CHARACTERISTICS: On stems, the fungus produced dark, shiny, round to oval, slightly protuberant sclerotium-like bodies (FIG. 2A). The flyspeck on stems did not damage the plants, but greatly reduced their ornamental and retail value. As a result, these fungi can cause significant economic losses to producers of these medicinal plants.

KOCH'S POSTULATES: The inoculated apples were examined after incubating for 1 month. All inoculated apples with the two isolates exhibited flyspeck signs similar to that on the original plant stems, although with a sparser density of the sclerotium-like bodies. Control apples did not show any flyspeck signs.

## Discussion

Based on phylogenetic analysis of the ITS region and morphological characters of the anamorph, we identified the two isolates as *Strelitziana africana*. This species was described from *Strelitzia* (Arzanlou & Crous 2006) and was previously known only from that host. This study is the first report of *S. africana* from medicinal plants. Currently there are only two species in *Strelitziana*, and no potential teleomorph connection is known. *Strelitziana africana*, the type species of this genus, has rhexolytic conidiation and conidial dimensions similar to *S. australiensis* Cheewangkoon & Crous (Cheewangkoon et al. 2009). However, *S. africana* lacks an apical mucilaginous appendage and chlamydospores and has obclavate conidia, making it easy to distinguish from *S. australiensis*.

Morphologically, our two isolates are similar to *Strelitziana africana*, though our isolates produced longer conidia and conidiophores ( $(12\text{--})35\text{--}70(100)\ \mu\text{m}$ ,  $3\text{--}20(50)\ \mu\text{m}$ ) than *S. africana* ( $(18\text{--})50\text{--}70(95)\ \mu\text{m}$ ,  $3\text{--}15(40)\ \mu\text{m}$ ). Furthermore, conidiophores and conidia of the Chinese isolates produced

more septa (0–4 in conidiophores, 3–10(–21) in conidia for these isolates, vs. 0–1(–5) in conidiophores, and 3–5(–10) in conidia of ex-type strains of *S. africana*). In ITS sequence analysis, our isolates and *S. africana* isolates from *Strelitzia* identified by Arzanlou & Crous (2006) fell within a single clade with 100% bootstrap support.

The results of Koch's postulates show that: 1) the fungi from medicinal plant can produce flyspeck signs on apple fruit; 2) medicinal plants may therefore act as reservoir hosts, providing inoculum for SBFS infestations on apple. Based on the ITS sequence analysis and morphological comparison, we identified the isolates as *Strelitziana africana*, which represent a new record for China.

### Acknowledgments

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## BOOK REVIEWS AND NOTICES

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## INTRODUCTION

A variety of topics forms the subject of the books reviewed in this instalment from updated versions of books on ascomycetes, Moroccan gilled mushrooms, and European marasmioid and collybioid species, to a Japanese treatment of several genera in the *Agaricales*, a fungal inventory of the Black Forest's swamps, mires, and bogs, an introduction to Alaskan cryptogams, and an overview of fungi in all their aspects. A list of newly published books to be included in upcoming BOOK REVIEWS AND NOTICES is given at the end.

## ASCOMYCETES

**Schimmelpilze und deren Bestimmung. 3. neu bearbeitete Aufl.** By L.E. Petrini & O. Petrini. 2010. J. Cramer in der Gebrüder Borntraeger Verlagsbuchhandlung, Johannesstraße 3A, 70176 Stuttgart, Germany. <mail@schweizerbart.de>. Pp. x + 170, figs 33. ISBN 978-3-443-50035-1. Price € 39.80.

This short textbook first appeared in the series BIBLIOTHECA MYCOLOGICA in 2002 (see MYCOTAXON 86: 480–481, 2003) and was reissued in that series in 2008 with only very minor changes (see MYCOTAXON 110: 511–512, 2009). The first obvious difference in this third edition is that it is not released as a part of the BIBLIOTHECA MYCOLOGICA series — it has attractive coloured front and back covers with photographs rather than the standard bright green of a BIBLIOTHECA MYCOLOGICA, something that will immediately make it more appealing to students. The book has also swelled by 26 pages, has five more figures, and I was personally gratified to see that the authors had acted

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<sup>1</sup> Books for consideration for coverage in this column should be mailed to the Book Review Editor at the address above. All unsigned entries are by the Book Review Editor.



on particular points raised in my review of the second edition. This was not just a matter of correcting author attributions, and inserting cited references missing from the “Literatur” section but entailed the adoption of more recent molecular classifications and references, including the demise of the category “deuteromycet” — although “Coelomycetes” and “Hyphomycetes” persist for pragmatic reasons as headings in the keys. The authors have in large measure vindicated my comment “that they could have produced a book that was more authoritative and reflected our current knowledge of mould fungi”. I was especially pleased to see that much of the increased length was due to new entries for additional genera, along with the re-arrangement and expansion of some of the photograph plates so as to include details of additional fungi. Unfortunately, the reproduction quality of some photographs taken from the previous edition is far from optimal (e.g., Abb. VIII.4). In addition, the short section on chemotaxonomic and molecular approaches to classification has been extended slightly; perhaps the section could be even longer in a fourth edition to explain in some detail the different molecular methods that can be employed in identification and the pitfalls of relying solely on sequence-comparisons. This textbook will now be even of more value to German-speaking students than the earlier editions. Indeed, those who bought the second edition should promptly discard it and buy the third!

My final comment is that I would like also to see this available in an English translation, especially as there is currently no really equivalent work in print. In particular, the plates showing different types of conidiogenesis in detail merit a much wider audience than they will receive hidden in a German text-book. I will be interested to see if this suggestion is taken up, and, if it is, I will be really pleased with the additional evidence that comments made in book reviews can have tangible results; the genre would then have been unequivocally vindicated.

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## GUIDEBOOK

**Common interior Alaska cryptogams. Fungi, lichenicolous fungi, lichenized fungi, slime molds, mosses, and liverworts.** By G.A. Laursen & R.D. Seppelt. 2009. University of Alaska Press, PO Box 756240, Fairbanks, AK 99775, U.S.A. <fyppress@uaf.edu> Pp. 256, plates 338, figs 113. ISBN 9781602230583. Price \$26.95.

The vast and highly varied landscapes of interior Alaska provide a home for numerous fungi and mosses and liverworts, to which a first introduction is given in this field guide.

An introduction to the area, well known because of Denali National Park, to the various groups of organisms treated in this book, and to mushroom hunting in these wild areas precedes the main part of the book in which mushrooms distributed over various groups such as gilled mushrooms, gasteromycetes and assorted ascomycetes, lichens and lichenicolous fungi, slime molds, and mosses and liverworts are described. A glossary, list of references, and an appendix in which mycological reagents are treated finish off the book.

Each species is represented with a small photo and, in the case of the mosses, also with line drawings. The fungal photos are not always sharp and in some cases overexposed, which makes identification hard. Keys to the species are not provided, and only a small selection of species made it into the book (e.g., 80 gilled fungi). This is the first guidebook I have seen in which lichenicolous fungi have been given a place, which is really a very nice asset. Some of the names are a little out of date (e.g. *Microcollybia* for what is now *Collybia* and *Collybia* for *Gymnopus*; *Rozites caperatus* for *Cortinarius caperatus*). There is no indication whether the descriptions and the photos are based on the same specimens or whether the specimens that got their photos in the book were conserved in a herbarium.

The copy I read has several misprinted pages, and I hope that this is not a widespread problem.

All in all, it is a nice first introduction to mushrooms, fungi, and mosses of the fascinating northwest.

#### AGARICALES AND OTHER GILLED MUSHROOMS

**Compléments à la Flore des champignons supérieurs du Maroc de G. Malençon et R. Bertault.** By J.-C. Maire, P.-A. Moreau, G. Robich (editors). 2009. Confédération européenne de mycologie méditerranéenne, Nice. Pp. 775, plates 58, figs 50. No ISBN number. Price ca. € 116.00.

The out-of-print work by Malençon & Bertault (1970, 1975) on Moroccan mushrooms is a classic example of well-executed, thorough flora work based on meticulous observations, fieldwork over a long period of time, and a sound insight in fungal taxonomy.

With a huge increase in interest in the mycoflora of the southern European Mediterranean countries, an update was deemed necessary. The CEMM (Confédération européenne de mycologie méditerranéenne) initiated the project of which this book is the result. It is not just a review of the original work with updated names — no, original material, collections, and water colours (many unpublished) that were kept at MPU were for the first time sent out to the contributors.

A total of 32 mycologists (amateur and professionals) from eight countries contributed, and the level of depth and manner of treatment was more or less left to the discretion of each contributor. There are pieces in French, Italian, Spanish, and English. The coordinator, Pierre-Arthur Moreau, regretted that it was not possible to include *Russula* and *Psathyrella* in the present work. During the research ten new taxa were discovered or names proposed by Malençon validated. Also a number of new combinations was necessary. For each species, the name originally used by Malençon & Bertault, the currently accepted or adopted name, lists of material examined, and comments are given. Original drawings and watercolours (both in colour and in black-and-white) from the notes made by Malençon, in some cases by the authors, illustrate the species.

For some groups a detailed account of all collections is given (e.g., for *Lactarius* sect. *Deliciosi* in the revision by Jorinde Nuytinck) while treatment is much less detailed for others, but overall the extensive discussions and notes are interesting and insightful. The genus *Hygrocybe* is included in two versions due to a small mistake by the coordinating editors.

Interesting is the find of a putative species of the genus *Cleistocybe*, a genus that was recently described (Ammirati et al. 2007) for veiled *Clitocybe*-like species. In general the names are up to date, although in some cases the authors adhere to some older generic concepts (e.g. *Stropholoma* instead of *Leratiomyces*; *Sericeomyces* instead of *Leucoagaricus*).

This is an extremely valuable work, contributing greatly to our knowledge of north African and Mediterranean taxa in general, and also a great example of a group effort to which many contributed.

Ammirati JF, Parker AD, Matheny PB. 2007. *Cleistocybe*, a new genus of *Agaricales*. *Mycoscience* 48: 282–289.

Malençon G, Bertault R. 1970. Flore des champignons supérieurs du Maroc. I. Travaux de l'Institut scientifique chérifien et de la Faculté des Sciences de Rabat, Série botanique et biologie végétale 32: 604 pp., 133 figs, 33 pl.

Malençon G, Bertault R. 1975. Flore des champignons supérieurs du Maroc. II. Travaux de l'Institut scientifique chérifien et de la Faculté des Sciences de Rabat, Série botanique et biologie végétale 33: 540 pp., 105 figs, 22 pl.

**Taxonomic studies on *Agaricales* of Hokkaido, Northern Japan, with special reference to *Melanoleuca*, *Oudemansiella*, *Xerula*, *Volvariella* and *Pluteus*.** By S. Takehashi, T. Hoshino & T. Kasuya. 2010. Non profit organization The forum of Fungi in northern Japan, Kanayama 1-3 10-3, Teine-ku, Sapporo, Hokkaido, 006-0041, Japan. <BXG05024@nifty.com>. Available from SANO Books, Sakae-machi 6-19, Aioi-city, Hyogo 678-0008, Japan, <e\_sano@d2.dion.ne.jp>. Pp. 145 + xiii, numerous plates, numerous figs. ISBN 978-4-9905010-0-6. Price ¥ 5.600.

Keys and descriptions with photos and line drawings for four genera of *Agaricales* from Hokkaido form the mainstay of this book assembled by the

“Forum of Fungi” in northern Japan, a group of mushroom enthusiasts who have been researching and documenting the fungi of this northernmost of the four main islands of Japan. Since the 1938 publication by Imai, there has not been a comprehensive update of this seminal work, until this initiative, which treats *Melanoleuca*, *Oudemansiella*, *Xerula*, *Volvariella*, and *Pluteus*. The taxonomic part, which is in Japanese and English, is interspersed with Japanese only sections, with descriptions of some of the collecting sites, fungal names used by the Ainu, fruiting phenology, Hokkaido in fungal epithets, fungi under snow cover, and a separate page on the morphology and ecology of *Melanoleuca verrucipes*.

A total of four *Melanoleuca*, three *Oudemansiella*, 13 *Xerula*, four *Volvariella*, and 28 *Pluteus* taxa are treated in detail. It is impressive that this volume tackles two difficult genera, *Pluteus* and *Melanoleuca*. Species that could not be identified are presented as “X” sp., and comments and comparisons with the literature etc. are given for each taxon. The descriptions are complete and the illustrations are adequate (including SEM photos of the spores for *Melanoleuca*) and clearly labeled as to which collections they reference.

It is a bit of a disappointment to discover that the name ‘idahoensis’ does not refer to a place in Japan, but to the western American state of Idaho!

This work will serve as a modern treatment for these genera for Japan, and as an example for other areas of the world.

Imai S. 1938. Studies on the *Agaricaceae* of Hokkaido. I. Journal of the faculty of Agriculture Hokkaido Imperial University 43: 1–378.

**A monograph of marasmioid and collybioid fungi in Europe.** By V. Antonín & M.E. Noordeloos, 2010. IHW-Verlag & Verlagsbuchhandlung, Postfach 1119, 85378 Eching, Germany <dr.schmid@ihwverlag.de>. Pp 480, figs 131, Plates 130. ISBN 978-3-930167-72-2. €139.00.

This is the updated version of the two earlier volumes on the marasmioid and gymnopoid fungi of Europe (Antonín & Noordeloos 1993, 1997); more colour photos, especially for the marasmioid species, recently described species, and new notes have been added. With almost 500 pages and 130 colour plates, the ‘monograph’ is a very impressive book indeed. To free space, the lists of examined specimens and excluded species are provided separately on a CD that comes with the book. An introduction to the group, overviews of past classifications, and phylogenetic placements inferred from phylogenetic DNA analyses are given in the introduction, together with an explanation of names selected (see below). Keys to the genera, groups, and species make it relatively easy to find a name for your specimens. Extensive descriptions of macroscopical and microscopical characters for each taxon, notes on ecology, occurrence, and

habitat, and comments on similar taxa and other interesting and necessary information are included.

By publishing now, the authors had to make many decisions on the classification, as the phylogeny of this group of species is in no way completely settled. For instance, just this spring, Hughes et al (2010) proposed the new genus *Connopus* for *Gymnopus acervatus*. The group of species treated in this book, once smugly accommodated in the white spored catch-all family *Tricholomataceae*, has been shown to fall into four families (*Tricholomataceae* in a strict sense, *Marasmiaceae*, *Omphalotaceae*, and *Physalacriaceae*). Various authors (e.g. Mata et al 2004, 2006; Wilson & Desjardin 2005) have studied different taxa in this group using different sets of molecular markers as a base for their phylogenetic hypotheses, resulting in a jig saw puzzle for which we still only have a small number of pieces, some of them fitting together, others representing different parts of the picture. It also does not help that the majority of the species of *Marasmius* and *Marasmiellus* is tropical. Antonín & Noordeloos, who did not perform molecular-phylogenetic analyses on the European taxa themselves, relied on work by others. They decided to treat *Setulipes* (*Marasmius androsaceus* et al) within *Gymnopus* {based primarily on work by Mata et al. (2006)} — still quite controversial, as up to now only a very few *Setulipes* species have been analyzed. *Micromphale* species are also included in *Gymnopus*, whereas in 1997 Antonín and Noordeloos placed them in *Marasmiellus*. *Marasmius*, *Gymnopus*, and *Marasmiellus* are recognized as being not monophyletic, with *Marasmius* even spread over two families. But as the dust has not yet settled on the phylogenies of these taxa, this is indeed a good compromise. *Gloiocephala*, the small gelatinized and almost lamella free species growing on grasses and the like, is again a separate genus, not at all close to the core group of *Marasmius*. *Mycetinis* accommodates the garlic smelling species formerly placed in *Marasmius*; but now *M. epidryas* is also in *Mycetinis*. It must be frustrating for users to know that more name changes will be imminent.

At the species level, the group of species around *Gymnopus dryophilus* definitely needs more work: Mata et al (2006) showed that *G. ocior* is restricted to North America, yet the name is still maintained here for a European species.

With a work of this size there are of course many details to attend to, and there the book does fall short. Species numbers in keys are occasionally off, the two parts of a lemma of a key may cite the same habitat, the list of substrate specialists taken from the 1993 book omits most of *Gymnopus*, a figure legend may still have the old genus name, and so on.

Nevertheless, these are minor comments on a very useful, well illustrated, very informative book.



- Antonín V, Noordeloos ME. 1993. A monograph of *Marasmius*, *Collybia* and related genera in Europe. Part 1: *Marasmius*, *Setulipes*, and *Marasmiellus*. Libri Botanici 8.
- Antonín V, Noordeloos ME. 1997. A monograph of *Marasmius*, *Collybia* and related genera in Europe. Part 1: *Collybia*, *Gymnopus*, *Rhodocollybia*, *Crinipellis*, *Chaetocalathus*, and additions to *Marasmiellus*. Libri Botanici 17.
- Hughes KW, Mather DA, Petersen RH. 2010. A new genus to accommodate *Gymnopus acervatus* (Agaricales). *Mycologia* doi:10.3852/09-318
- Mata JL, Hughes KW, Petersen RH. 2004. The phylogenetic position of *Marasmiellus juniperinus*. *Mycoscience* 45: 214–221.
- Mata JL, Hughes KW, Petersen RH. 2006. An investigation of /omphalotaceae (*Fungi*: Euagarics) with emphasis of the genus *Gymnopus*. *Sydowia* 58: 191–289.
- Wilson AW, Desjardin DE. 2005. Phylogenetic relationships in the gymnopoid and marasmioid fungi (*Basidiomycetes*, euagarics clade). *Mycologia* 97: 667–679.

## FUNGAL INVENTORIES

**Die Funga der Moore des Hochschwarzwaldes.** Ergebnisse einer Langzeitstudie. By D. Laber. 2010. Beiheft zur Zeitschrift für Mykologie Band 11. Deutsche Gesellschaft für Mykologie, Postfach 700447, 81304 München, Germany. <schatzmeister@dgfm-ev.de>. Pp 208, figs 43, plates 84. Price € 25.

Over the course of 34 years, 578 forays into different types of moors, swamps, bogs, brooksides, and other watery habitats in the higher ranges of the Black Forest were undertaken by the author and her husband. The results of all this work are reflected in this excellent publication. A thorough introduction, with detailed information on the visited areas, their vegetation, and their fungi is followed by a checklist of all 671 fungal species identified and detailed descriptions of selected interesting species. The author focused on the more conspicuous basidiomycetes and ascomycetes, leaving to one side crust fungi and the ascomycetes that form small fruitbodies. The area and its fungi stand out because of the granite and gneiss bedrock, which complicates comparison with the mycoflora of other similar European habitats situated on limestone (Favre 1948 for the Jura, Moreau 2002 for the French northern Alps, Einhellinger 1976 & 1977 for Bavarian swamps and bogs). One finding in the present work is that species regarded as northern and occurring in Scandinavia are also at home in this montane area further south. I am very impressed by the fact that the names are very up-to-date; it is clear that a huge effort has been put into using the proper nomenclature, an extraordinarily difficult task that demands a large library.

The checklist part gives for each taxon the exact habitats, the number of finds per location, the fruiting period, the highest altitude found, and a picture reference. Full descriptions are complemented by line drawings and discussions of the finds.

This work is important in a number of ways. It gives the mycoflora of one area at one point in time, which is great in respect to possible changes that will take place; currently the area still gets a high amount of precipitation, but this might change in the future. Such a comprehensive and illustrated overview of the fungi in one habitat type in one region can help identify species in other similar habitats. An extremely good example of what dedicated research over a long period can achieve, the 'Funga' also demonstrates that one need not be a professional mycologist to contribute. Similar research, but not as long-running, was done by a team of mycologists on Vancouver Island (Canada) (e.g. Roberts et al. 2004), but the longevity and thoroughness of the German study make it stand out. In short, the Laber work is highly recommended, not only for those interested in central European mycoflora but also for everybody interested in doing fungal surveys themselves.

Einhellinger A. 1976. Die Pilzen in primären und sekundären Pflanzengesellschaften oberbayerischer Moore, Teil 1. Ber. Bayer. Bot. Ges. 47: 75–149.

Einhellinger A. 1977. Die Pilzen in primären und sekundären Pflanzengesellschaften oberbayerischer Moore, Teil 2. Ber. Bayer. Bot. Ges. 48: 61–146.

Favre J. 1948. Les associations fongiques des haut marais jurassiens. Beiträge zur Kryptogamenflora der Schweiz 10: 1–228.

Moreau P-A 2002. Analyse écologique et patrimoniale des champignons supérieurs dans les tourbières des Alpes du Nord. Thesis Université de Savoie. 336 pp.

Roberts C, Ceska O, Kroeger P, Kendrick B, 2004. Macrofungi from six habitats over five years in Clayoquot Sound, Vancouver Island. Canadian Journal of Botany 82: 1518–1538.

## FUNGI IN GENERAL

**The kingdom *Fungi*. The biology of mushrooms, molds and lichens.** By S.L. Stephenson. 2010. Timber Press, 133 SW 2nd Avenue #450, Portland, OR 97204, U.S.A. <info@timberpress.com>. Pp. 328, plates 124. ISBN 978-0-88192-891-4. Price \$34.95, £ 20.00.

Not a text book, not a collection of fungal stories, but an introduction to the world of fungi for lay persons and amateur mycologists – that is what this book boils down to.

After an introduction to the subject, various groups of fungi, not necessarily taxonomic units, are treated. Aquatic fungi first, followed by terrestrial fungi divided into subgroups (ascomycetes and zygomycetes, truffles and their kin, gilled fungi and other basidiomycetes, lichens and slime molds). Chapters on the roles of fungi in the environment, interactions between fungi and humans and other animals, and fossil fungi, plus a glossary and a list of references make this book complete. Two sets of colour photos, a total of 124 plates, illustrate it quite nicely. There are neither diagrams of life cycles and such nor phylogenetic trees to clarify the concepts given in the text.

From the list of chapters it should be clear that also some non-fungi such as the water molds and slime molds are treated, as they look and behave like fungi and have traditionally been studied by mycologists. The author indicates that many of the old categories and classifications do not hold up in the age in which phylogenetic methods to compare DNA sequences have revolutionized fungal classifications, but he still uses old terms such as gasteromycetes and aphylophorales. That is a missed chance, in my opinion, for such a book is an excellent place to introduce amateur mycologists to new insights.

THE KINGDOM *FUNGI* contains enough interesting tidbits and fascinating mycological oddities to entice the reader, and it might pave the road to other more comprehensive books.

### BOOK ANNOUNCEMENTS

**Les myxomycètes.** By M. Poulain, M. Meyer & J. Bozonnet, 2010. FMBDS, 8, Avenue de la Plaine, 74000 Annecy, France, <philippecattin74@orange.fr>. 2 vols, Plates 546. € 80 (before Oct 2010) € 120.

**FoodMold: an interactive CD guide to foodborne fungi.** By J.I. Pitt, E. Rico-Munoz & E.S. Johnson, 2009. BCN Research Laboratories, 2491 Stock Creek Blvd, Rockford, TN 37853, U.S.A. foodmold@bcnlabs.com. \$ 340.

**Fascinating microfungi (hyphomycetes) of Western Ghats, India.** By D.J. Bhat, 2010. Broadway Book Centre. Pp xii, 222, figs 127. \$80.00 (includes shipping).

**Systematics of *Calonectria*: a genus of root, shoot and foliar pathogens.** By L. Lorenzo, P.W. Crous, B.D. Wingfield & M.J. Wingfield, 2010. Studies in Mycology 66. CBS Fungal Biodiversity Centre, Uppsalalaan 8, 3584 CT, Utrecht, the Netherlands, <http://shop.fungalbiodiversitycentre.com> Pp 71 € 40.



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## Fungal nomenclature

### 1. The IMC9 Edinburgh Nomenclature Sessions

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Three successive groundbreaking two-hour long nomenclatural sessions were held August 3–5, 2010, during this summer's International Mycological Congress (IMC9) in Edinburgh, Scotland. Convener/Rapporteur David Hawksworth (Spain/UK), who supervised preparation of the IMC9 nomenclatural booklet + questionnaire, was assisted by Chair Ron Petersen (USA), Vice-Chair Scott Redhead (Canada), Nomenclature Committee for Fungi (NCF) Secretary Lorelei Norvell (USA), and Advisor & International Botanical Congress Rapporteur-général John McNeill (UK). IMC delegates attending each day's session voted on nomenclatural proposals to recommend actions to next year's International Botanical Congress (IBC) Nomenclature Section in Melbourne. Attendance was relatively high, particularly in view of the conflict caused by scheduling the three nomenclature and three (of four) poster sessions for the same 2–4 pm time periods. As each poster session presented authors and posters for only one day, this was an unfortunate conflict that influenced attendance numbers at the nomenclatural sessions. However, the questionnaires, distributed to all IMC9 delegates for return to the registration desk by the end of the Congress, permitted each delegate a chance to express an opinion, even if unable to attend any or all of the Nomenclature Sessions.



Originally the entire proceedings, which proved to be lively, informative, and often amusing, were to be recorded. Due to an unfortunate communications failure, no recordings survive. The overly brief summary below has therefore been extracted from secretarial notes, the nomenclature booklet, and the returned questionnaires.

## Background

When initially formed in 1971, the International Mycological Association (IMA) established a Nomenclature Secretariat to address issues of concern to mycologists. This led to a series of proposals on starting points and other matters that were adopted by the International Botanical Congress in Sydney in 1981, after which it was disbanded, having completed its tasks. Since that time, discussions of nomenclatural issues at IMCs have been confined to occasional debates on particular topical issues. However, at IMC8 in Cairns in 2006, some delegates spoke strongly in favour over a separate Code for fungi. Subsequently, proposals that could fundamentally change aspects of fungal nomenclature have been published; these are to be voted on at the forthcoming International Botanical Congress (IBC) in Melbourne in July, 2011. As IBCs occur only every six years, and decisions made there generally come into force 1-2 years later, any issues not decided in 2011 would have to wait until 2018 or 2019 to be implemented. The Nomenclature Sessions at IMC9 were convened to (1) enable a broad spectrum of mycologists to express their views on a wide range of topics and also to vote on proposals already made and (2) establish that IMCs can incorporate effective Nomenclatural Sessions.

## Session 1: Governance of fungal nomenclature

Approximately 100 delegates attended the first session convened by Hawksworth at 2 pm on August 3. After Chair Petersen set forth the 'rules of engagement' for audience participation during all sessions, two introductory background presentations were given. Vincent Demoulin (Belgium, Chairman of the Committee for Fungi) spoke in defense of retaining governance of fungi within the Botanical Code and Hawksworth reported on the progress being made toward one unified code for all organisms. (See Appendix 1, below.)

The floor was then opened to discussion of the formal proposals for the governance of fungal nomenclature, the composition of the Nomenclature Committee for Fungi, and a (very) brief discussion of the proposed exclusion of *Microsporidia* from the ICBN. At the close of the two-hour session, those remaining in the auditorium were polled as to their preferences, summarized as follows:

PROPS. 016–020 (see MYCOTAXON 108: 1–4) all passed. Votes were actually counted for the first two proposals: both PROP. 016 (to amend the current

Botanical Code to establish more clearly that it covers fungi, including changing the name to the “International Code of Botanical and Mycological Nomenclature”) & PROP. 017 (to replace “plants” by “plant(s) or fungus/fungi” throughout) passed with 87 yes and 4 no votes. Thereafter, due to time pressures, only the ‘no’ votes (out of 91 total) were counted, with 3 voting against Prop. 018 (to provide for the election of the Permanent Nomenclature Committee for Fungi by an International Mycological Congress), 3 voting against Prop. 019 (to relegate decision-making on proposals relating solely to organisms treated as fungi to an IMC), and 1 against PROP. 020 (to insert a new Div. III.5 requiring the presence of the Secretary for the Committee for Fungi or Committee alternate on the Editorial Committee).

Unanimous support was given to retaining the current members of the COMMITTEE FOR FUNGI until the 2014 IMC10 in Bangkok, provided that the 2011 International Botanical Congress in Melbourne accepts the fungal governance proposals above.

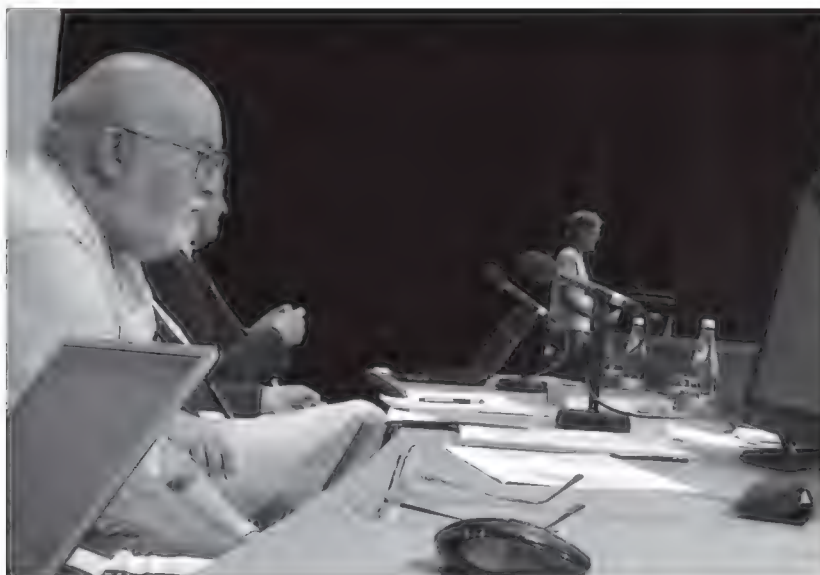
PROPS. 048–051 (to exclude the governance of the phylum *Microsporidia* from the Code; see MYCOTAXON 108: 505–507) passed with only one dissenting vote, but as the vote was held as delegates were leaving the session, it may not accurately reflect the wishes of the majority. Demoulin has since submitted PROP. 190 to limit Art. 45.4 (p. 514, this volume).

## **Session 2: Mandatory pre-publication deposit in a nomenclatural repository, electronic publication, type cultures, and illustrations**

After opening introductions, Paul Kirk (UK) provided an overview of the current strides made in data-basing taxonomic names of all organisms worldwide. (See Appendix 1, below.)

A fluctuating audience (estimated at 97 total for the 2-hour session) discussed at length and eventually recommended PROPS. 117–119 (see MYCOTAXON 111: 514–519). PROP. 117 (to require deposition of names and required nomenclatural information in a recognized repository (such as MycoBank) for valid publication) received 58 yes, 5 no, and 1 abstaining votes. PROPS. 118 (to recommend deposit of minimal information elements, accession identifiers, and bibliographical details for valid publication) and 119 (to require citation of a repository identifier for valid publication) received almost universal support, with 1 and 2 abstentions respectively. Kirk also announced that it would be possible to deposit names with the Index Fungorum, although the mechanism (still in progress) was not detailed.

An informal poll showed no clear consensus for or against valid electronic publication of names. PROP. 138 (which seeks to add Rec. 8B.3, including the phrase ‘permanently preserved in a metabolically inactive state’ or its equivalent



Chairman Ron dominates the auditorium while Paul Kirk explains nomenclatural databases on August 4.

when designating a culture as a type) likewise showed no clear consensus with the majority abstaining.

The session concluded with a second informal poll (showing 4 for, 25 against, and the majority abstaining) regarding the addition of illustrations as a requirement for valid publication.

### **Session 3: Moving to one name for one fungus & ending the requirement of Latin diagnoses for valid publication**

Approximately 145 delegates attended the final (and most controversial) “Article 59” session on August 5. Background on attempts to modify dual nomenclature was provided by Redhead (Secretary for the Special Committee on Names of Fungi with a Pleomorphic Life History), followed by a presentation by Walter Gams (Netherlands), who spoke on the limitations of “teleotypifying” fungal names according to Art. 59.7. (See also Appendix 1, below.)

Emotions ran high in this session, and discussion was lively, entertaining, lengthy — and inconclusive. No formal proposals were before the Session, so no vote was scheduled on Art. 59. It was assumed that Congress participants would mark their opinions on their questionnaires.

Due to the lengthy Art. 59 debate, the scheduled discussion and vote on whether to end the requirement of a Latin diagnosis for the valid publication of scientific names (also to be considered in 2011 at Melbourne) became a side issue. Entrants crowding the doors for the next scheduled mycological session dictated Chair Petersen’s decree for adjournment, which drowned out the plaintive cry from the back of the hall, “Why can’t we vote to abolish Latin?” and a call to hold a vote on Art. 59.

### Final resolution approved by the General Assembly — and a note of caution

At the close of the first Nomenclature Session, 103 questionnaires had already been returned. By the evening of the final session, Hawksworth and Norvell had tabulated 167 results and identified three clear preferences for presentation to the delegates during the IMC9 closing ceremonies on August 6. The General Assembly voted by acclamation to approve the resolution below:

This General Assembly of the IMA endorses the decisions of the Nomenclature Session convened during IMC9 with respect to

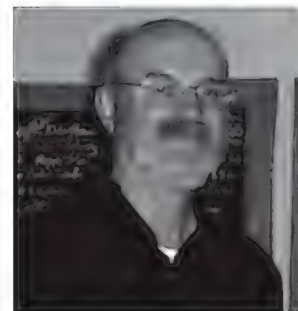
- the transference of the governance of the nomenclature of fungi from the International Botanical to International Mycological Congresses,
- the mandatory pre-publication deposit of nomenclatural information in a recognized depository for the valid publication of new fungal names,
- the acceptability of English as an alternative to Latin in the valid publication of fungal names,

and requests the permanent Nomenclature Committee for Fungi, the special Committee on the names of Pleomorphic Fungi, the International Commission on the Taxonomy of Fungi, and the next International Botanical Congress to take note of the results of the questionnaire completed by delegates of IMC9.

In summary, we must emphasize that these are recommendations and not approved changes. Currently fungal names are still governed by the International Code of Botanical Nomenclature, and — until changed — a Latin description or diagnosis is still required, as are other established requirements for valid publication as set forth in the current International Code of Botanical



Petersen, Hawksworth, and IBC Rapporteur-général McNeill await the Art. 59 'discussions' on August 5.



Vice-Chair (& photographer) Redhead post-session



Nomenclature (McNeill & al. 2006). Nonetheless the interest shown in nomenclature at IMC9 was gratifying, and we are optimistic that many of the innovations supported by most mycologists will be made.

### **Appendix 1: IMC9 Nomenclature Session presentation abstracts**

**FEWER NOMENCLATRURAL CODES, NOT MORE, IS WHAT WE NEED (Demoulin):** At the first IMC (Exeter, 1971) the idea of a nomenclature code especially for fungi was discussed and a nomenclature committee was created under the auspices of the IMA. This committee reported at the 2nd IMC in Tampa, Fla. 1977. At that congress, the idea of a mycological code was abandoned in favour of more involvement by mycologists in the elaboration of the Botanical Code, which has ruled the nomenclature of fungi since its origin. A consequence was the important change in the starting point system adopted at the 13th International Botanical Congress (Sydney, 1981).

**PROGRESS TOWARDS A BIOCODE (Hawksworth):** In October 2009, the General Assembly of the International Union of Biological Sciences (IUBS) decided to re-activate the initiative to produce a unified Code of nomenclature for all organisms, by updating the DRAFT BIOCODE (1997). This is being taken forward by the International Committee for Bionomenclature of the IUBS/IUMS (International Union of Microbiological Societies). The need for, and route towards, a revised and agreed BIOCODE is reviewed as a background to the Session's deliberations.

**A WEB OF DATA FOR FUNGAL BIOLOGY RESEARCH— THE REGISTRATION QUESTION (Kirk):** Why do we give names to fungi? It's a simple question with a simple answer - to allow us to effectively communicate about the fungi, for the name is the link to all that is known about the organism. But in this answer the word 'us' is already of secondary importance. The web is the primary means of communication today and increasingly that means computer to computer communication. In addition, the current version of the web - a web of information - is rapidly being replaced by a web of data (the Semantic Web, especially Linked Data using RDF triples of entity-attribute-value) which will allow more rapid (real time) advances in synthesis, analysis, hypothesis, etc. The founder of the web Tim Berners-Lee, amongst others, is pushing for this to happen and we can be part of this effort. This short presentation will describe how name registration can operate, how associated data can be made available, what the barriers are, and how it all fits into existing and developing major global initiatives. It will indicate how fungal taxonomist and nomenclaturalists can be part of this with respect to the names we give to fungi.

**HOW DO MYCOLOGISTS WISH TO TREAT NAMES BASED ON ANAMORPHS? (Redhead):** Fungal nomenclature dates back to Linnaeus (1753) when the use of microscopes was limited and the existence of sexual life cycles amongst them was unknown. Nearly 200 years later (1935) mycologists realized they had been naming different



parts of fungal life-cycles as new species or genera, and formalized nomenclature rules giving priority to names for pleomorphic fungi based upon perfect states. Exceptions and refinements were instituted in 1950 and continue today. Many fungi only produce anamorphs, many generic names are based upon anamorphs, and many fungi are better known under anamorph names. However, complications in merging and then prioritizing names have created a nightmare situation that has divided the mycological community and now acts as a roadblock. Proposals to block the deliberate generation of alternative names and smooth the transition to normal nomenclature were partially approved for Article 59 in the International Code of Botanical Nomenclature (2006) while remaining issues were referred to a Special Committee by the IBC. After >4 years this Committee was unable to reach consensus upon changes. Some mycologists have decided to ignore existing rules or to take nomenclatural risks. Genetic sequence phylogenetic analyses have revealed many new relationships leading to binomial recombinations and even a PHYLOCODE. Having reached an impasse it can be asked if mycologists wish to eliminate dual nomenclature? If the answer is yes, it may be asked how to resolve conflicts, and then to create a process or body capable of dealing with such conflicts.

**TELEOTYPIIFICATION OF FUNGAL NAMES AND ITS LIMITATIONS (Gams):** This presentation was submitted without a formal abstract and too late to be included in the printed program. Gams discussed the effects of ‘teleotypification,’ which permits — after a teleomorph discovered for a fungus previously known only as an anamorph (and for which there is no existing legitimate name for the holomorph) — designation of an epitype exhibiting the teleomorph stage for the hitherto anamorphic name, even when there is no hint of the teleomorph in the protologue of that name. Several examples were forwarded to show that teleotypification is not the same as ordinary epitypification. For further information, see Props. (172–174), p. 513, this volume.



Rapporteur David, José Dianese, and Secretary Lorelei tabulate questionnaire responses in the EICC registration hall on August 3.

**Appendix 2: IMC9 Nomenclature questionnaire results**

From August 1–10, IMC9 delegates returned questionnaires in which they were to circle a Y (yes) or N (no) to 24 questions on 4 topics. We discovered during our first tabulation that one number (#19) appeared twice, bringing the actual number of questions to 25, and have renumbered the text below accordingly. Of the 174 questionnaires received, 7 were declared ‘spoiled’ as the respondents had placed an X over an option so that we could not determine whether agreement or rejection was intended. Both raw numbers and majority percentages are shown. We note that protocols followed at the 2005 International Botanical Congress in Vienna with respect to the preliminary mail-in ballots decreed that propoals receiving 60% or higher support merited further discussion by the attending Nomenclature Section, while 75% support virtually ensured passage for all but the most controversial proposals. In the results reported below, opinions showing 60% (or greater) support are highlighted in bold.

**A. CODES OF NOMENCLATURE**

(Fungal names are now governed by the International Code of Botanical Nomenclature)

- 1 One code for the future nomenclature of all organism names would be ideal  
y-72 n-71 ... 50% (TIE)
- 2 Fungi should continue to be covered under the Botanical Code (ICBN)  
y-54 n-76 ..... 58% NO
- 3 Fungi should continue to be covered under the ICBN provided it is renamed  
the “Botanical and Mycological Code” y-97 n-40 ..... 71% YES
- 4 Fungi should be covered by a separate mycological Code (ICMN)  
y-51 n-91 ..... 61% NO
- 5 Under either ICBN or ICMN, decisions on fungal nomenclature should be  
voted at an International Mycological Congress (and not an International  
Botanical Congress), guided by a secure advanced web publication  
and mail/email votes y-133 n-21 .... 86% YES

**B. LANGUAGE REQUIREMENTS FOR VALID PUBLICATION OF NAMES**

- 6 Latin diagnoses/descriptions should continue to be required  
y-49 n-91 ..... 65% NO
- 7 English diagnoses/descriptions rather than Latin should be required  
y-69 n-69 ... 50% (TIE)
- 8 Either Latin or English diagnoses/descriptions should be required  
y-88 n-56 ..... 61% YES
- 9 Diagnoses/descriptions in any language should be permitted  
y-4 n-135 ..... 97% NO

**C. NOMENCLATRURAL INFORMATION DATABASING**

- 10 Deposition of key nomenclatural information in one or more approved  
depositories (e.g. MycoBank) should be made mandatory for the  
valid publication of new fungal names y-134 n-21 .... 86% YES
- 11 Historic names not included in *Index Fungorum* (after a set date) should  
no longer be treated as validly published y-55 n-68 ..... 55% NO
- 12 Deposited names should be automatically protected against any  
unlisted names after a date to be agreed y-90 n-39 ..... 70% YES
- 13 An accurate and free list should be prepared of names in use or  
available for use y-126 n-19 .... 87% YES

- 14 Names with key information deposited (e.g. in MycoBank) should be automatically available provided other Code requirements are met y-105 n-22 ... 83% YES
- 15 Electronic on-line only publication should be accepted without restriction y-24 n-126 .... 84% NO
- 16 Electronic on-line only publication should be accepted only when key nomenclatural information has been deposited (e.g. in MycoBank) y-113 n-36 .... 76% YES
- 17 For journals publishing online and printed copies, the dates of names should be those when the works are available in final form on-line y-101 n-40 .... 72% YES
- 18 For journals publishing online and printed copies, the dates of names should be those when the works are distributed in printed form y-63 n-73 ..... 54% NO
- 19 Special Group Committees should be empowered to create lists of acceptable and rejected names in particular groups (e.g. *Fusarium*, *Trichocomaceae*, yeasts) y-102 n-31 .... 77% YES

#### D. NAMES FOR PLEOMORPHIC FUNGI (ANAMORPHS, TELEOMORPHS)

- 20 The established system allowing dual nomenclature for anamorphs and teleomorphs should continue via Art. 59 y-67 n-71 ..... 51% NO
- 21 Article 59 should revert back to its status prior to changes in the 2006 Vienna Code, i.e. keeping separate anamorph and teleomorph names y-43 n-82 ..... 66% NO
- 22 A system of progressively establishing one name for each fungus should be enacted via modification of existing Articles (e.g. Art. 59) y-101 n-38 .... 73% YES
- 23 The historical practice of allowing valid names for different morphs of a species should be prohibited in the future via modification of existing Articles y-74 n-45 ..... 62% YES
- 24 The ability to select a “teleotype” (a type of epitypification) with a sexual state for a fungus previously only known in the asexual state should be continued y-88 n-31 ..... 74% YES
- 25 Article 59 (that permits the dual system) should be deleted provided other changes ensure this would not retroactively invalidate existing names y-66 n-47 ..... 58% YES

### Acknowledgments

We thank John McNeill (Royal Botanic Garden Edinburgh) for his perennially wise counsel and cheerful guidance. We further thank special presenters Vincent Demoulin, Paul Kirk, and Walter Gams; José Dianese (Brazil) for assisting in tabulating questionnaire results on August 3; and all those who participated in the nomenclatural discussions and/or completed questionnaires at IMC9 Edinburgh.

## 2. Proposals to conserve or reject fungal names

COMPILED BY LORELEI NORVELL

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**Abstract** — Formal proposals to conserve or protect fungal names are published in *TAXON* and considered by the IAPT permanent Nomenclature Committee for Fungi, which recommends conservation or rejection to the General Committee. The recently published Prop. 1945 (to conserve the name *Thelephora comedens* with a conserved type) is summarized and other proposals still under discussion by the Committee are listed. The complete text of all formal nomenclatural proposals is available on the internet at <[www.ingentaconnect.com/content/iapt/tax](http://www.ingentaconnect.com/content/iapt/tax)>. Those wishing to comment on a proposal still under consideration by the Nomenclature Committee for Fungi are invited to contact Secretary Norvell.

### Recently published

(1945) PROPOSAL TO CONSERVE the name *Thelephora comedens* (*Vuilleminia comedens*) with a conserved type (*Basidiomycota*). [Masoomeh Ghobad-Nejhad & Nils Hallenberg. 2010. *TAXON* 59(4): 1277–1278.]

**SUMMARY:** The name *Thelephora comedens* Nees : Fries is currently applied to a “basidiomycetous corticioid fungus presently known as *Vuilleminia comedens* (Nees : Fr.) Maire, which serves as type for the genus *Vuilleminia* Maire.” The epithet *comedens* is typified by a color drawing of a specimen in UPS that appears not to have been examined by anyone (including Fries) since publication of *T. comedens* in 1816–1817; the authors are unaware of any other Fries or Nees specimens representing *T. comedens* and regard the UPS specimen as the only extant material. The specimen does not conform to the current concept of *V. comedens* but represents a *Hyphoderma*. To preserve the stability of the species concept with the name, the authors propose as conserved type a specimen collected by Petrak from *Quercus*, which they regard to represent *T. comedens* as currently recognized.

### Other conservation proposals

(Committee for Fungi vote in progress)

PROP. 1769, to conserve the name *Cortinarius speciosissimus* against *C. rubellus*, *C. orellanoides*, and *C. rainierensis* (*Basidiomycota*).

PROP. 1810, to conserve the name *Hemipholiota* against *Nemecomyces* (*Agaricales*, *Basidiomycota*).

PROP. 1828, to conserve *Aspicilia aquatica* against *Lichen mazarinus* (*Ascomycota*: *Pertusariales*: *Megasporaceae*).

PROP. 1829, to reject the name *Verrucaria thelostoma*.

PROP. 1830, to reject the name *Pyrenula umbonata* (lichenized *Ascomycota*).

PROP. 1831, to conserve the name *Mixia* against *Phytoceratiomyxa* (*Basidiomycota*).

PROP. 1852, to conserve the name *Olivea tectonae* (T.S. Ramakr. & K. Ramakr.) R.L. Mulder against *Olivea tectonae* (Racib.) Thirum. (*Basidiomycota*).

PROP. 1861, to conserve the name *Aspicilia farinosa* (*Ascomycota*: *Pertusariales*: *Megasporaceae*) with a conserved type.

- PROP. 1862, to conserve the name *Psoroma versicolor* (*Degeliella versicolor*) against *Psoroma subdescendens* (lichenized Ascomycota, Pannariaceae).
- PROP. 1863, to conserve the name *Craterellus cinereus* (Pers. : Fr.) Donk with a conserved type against *Craterellus cinereus* Pers. (Basidiomycota).
- PROP. 1888, to conserve the name *Glomus* (*Fungi*, *Glomeromycota*, *Glomerales*) as being of neuter gender.
- PROP. 1896, to conserve the name *Lichen lichenoides* (*Leptogium lichenoides*) against *Lichen tremelloides* and *L. tremella* (lichenized Ascomycota).
- PROP. 1897, Proposal to reject the name *Lecidea epiploica* (lichenized Ascomycota).
- PROP. 1898, to conserve *Stirtonia* A.L. Sm. (lichenized Ascomycota, Arthoniales) against *Stirtonia* R. Gr. bis (Bryophyta, Dicranales).
- PROP. 1899, to conserve the name *Hebeloma cylindrosporum* against *Hebeloma angustispermum* (Basidiomycota).
- PROP. 1918, to conserve the name *Dermatocarpon* (*Placopyrenium*) *bucekii* against *Placidium steineri* (lichenized Ascomycota, Verrucariaceae).
- PROP. 1919, to conserve *Lactarius* (Basidiomycota) with a conserved type.
- PROP. 1926, to conserve *Cladia* against *Heterodea* (Ascomycota).
- PROP. 1927, to conserve the name *Agaricus rachodes* (Basidiomycota) with that spelling.

### 3. Proposals to amend the Code

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**Abstract** — Current proposals to amend the International Code of Botanical Nomenclature will soon be placed on a ballot to be distributed to all members of the International Association of Plant Taxonomists, scheduled for return prior to the 2011 International Botanical Congress in Melbourne, Australia. Summaries of recently published proposals of particular interest to mycologists are given for Props. 172–174 (to amend teleotypification procedures set forth in Art. 59.7), Prps. 183–184 (to require deposition of information concerning typification of fungal taxa), and Props. 185–190 (to amend Arts. 15, 36, and 45). Previous proposals also affecting fungal nomenclature are also listed. The complete text of all proposals is available for free download at <[www.ingentaconnect.com/content/iapt/tax](http://www.ingentaconnect.com/content/iapt/tax)>.

#### Recently published proposals

(Committee for Fungi vote in progress)

(172–174) Three proposals to amend Article 59 of the CODE concerning teleotypification of fungal names. Proposed by Walter Gams, Walter M. Jaklitsch, Roland Kirschner & Martina Réblová. TAXON 59(4): 1297.

SUMMARY: Three proposals to clarify the effect of teleotypification are provided. Prop. 172 proposes to eliminate Art. 59.7, thereby returning Art. 59 to the pre-Vienna situation. The alternate, Prop. 173, would modify the current Art. 59.7 so as to avoid including taxa with teleomorph-typified names within otherwise entirely anamorphic genera. If Prop. 173 is enacted, Prop. 174 would add Recommendation 59A.4, which specifies that newly discovered anamorphs should only be classified under teleomorph-



typified generic names when no suitable anamorph-typified generic names are available. Prop. 174 further specifies that a subsequent discovery of a teleomorph will require epitypification by a specimen exhibiting the teleomorph.

(183–184) Proposals to require deposition of information concerning typification of names of fungal taxa, with an associated Recommendation. Walter Gams. 2010. *TAXON* 59(5): 1626–1627.

**SUMMARY:** — Anticipating probable acceptance of Props. 117–119 to require deposition of nomenclatural information for valid publication of newly introduced fungal taxonomic names effective January 1, 2013, under PROP. (183) Gams would add a clause to Art. 7.10 that would make deposition in a recognized repository compulsory for effective typification from 2013 onwards.

PROP. (184) would insert new Recommendation 37bisA.2 (with appropriate cross-references) to encourage anyone publishing choices for names of fungal organisms to record the choice of name, orthography, or gender in a recognized repository, citing this and its record number in the place of effective publication.

(185–190) Proposals to amend Articles 15, 36, and 45. Vincent Demoulin. 2010. *TAXON* 59(5): 1627–1628.

**SUMMARY:** These five proposals were prompted by discussion in the Nomenclature Committee for Fungi and in the IMC9 Nomenclature sessions. PROP. 185 would insert into Art. 15.1 the sentence, “The spelling used by a sanctioning author is treated as conserved, except if it is to be corrected or standardized under Art. 60” and would instruct the Editorial Committee to insert an example to clarify what is meant by sanctioning. PROPS. 186–189 would modify Art. 36 to permit the use of a Latin OR English diagnosis, as is now permitted for fossil nomenclature under the current CODE. Prop. 190 seeks to limit Art. 45.4 to the first sentence, to be reworded as, “If a taxon is treated as belonging to the algae or fungi, any of its names need satisfy only the requirements of the non-botanical code that the author was using for status equivalent to valid publication under the present CODE (but see Art. 54 regarding homonymy.” A new Art. 45.5 is proposed to clarify that authors who regard organisms as representing fungi must also follow the CODE, which governs fungi, and not some other non-botanical code. This modification would thus make Art. 45 applicable to groups similar to the *Microsporidia* but which are not covered by Props. 48–51 (see below).

### **Other proposals to amend the CODE affecting fungi**

(Committee for Fungi vote in progress; see also IMC9 Nomenclature Session summary, this volume, pp. 503–509)

PROPS. (016–020), to amend the CODE to make clear that it covers the nomenclature of fungi, and to modify its governance with respect to names of organisms treated as fungi.

PROPS. (048–051), to exclude the phylum *Microsporidia* from the CODE.

PROPS. 117–119, to make deposition of nomenclatural information for all newly introduced names of fungal taxa a prerequisite for valid publication.

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- Aschersonia fusispora* Jun Z. Qiu, C.Y. Sun & Xiong Guan, p. 82
- Athelopsis parvispora* Avneet P. Singh, Dhingra & J. Kaur, p. 327
- Bionectria vesiculosa* J. Luo & W.Y. Zhuang, p. 245
- Ceratocystiopsis minuta* (Siemaszko) H.P. Upadhyay & W.B. Kendr. 1975  
(lectotypified), p. 13; (epitypified), p. 14
- Chaetospermum setosum* Rajeshkumar, S.K. Singh & P.N. Singh, p. 398
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- Leucoagaricus dacrytus* Vellinga, p. 74
- Marasmius asiaticus* Mešić & Tkalčec, p. 283  
= *Marasmius distantifolius* Y.S. Tan & Desjardin 2009, non (Murrill) Murrill 1915
- Marasmius canalipes* Tkalčec & Mešić, p. 284  
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= *Marasmius parvulus* Manim. & Leelav. 1987, non Berk. & M.A. Curtis 1858
- Marasmius lilacinitinctus* Mešić & Tkalčec, p. 284  
= *Marasmius lilacinus* (Coker & Beardslee) Singer 1951 ("1949"), non Henn. 1896
- Marasmius masseei* Tkalčec & Mešić, p. 284  
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- Marasmius neotropicus* Mešić & Tkalčec, p. 285  
= *Marasmius asemus* Singer 1989, non (Fr. : Fr.). P. Karst. 1889
- Monodictys macrospora* H.Q. Pan & T.Y. Zhang, p. 259
- Neobulgaria alba* P.R. Johnst., D.C. Park & M.A. Manning, p. 388

- Oudemansiella macracantha* Singer 1962 (lectotypified), p. 122  
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p.231, Abstract, line 3, for: <i>E. pallidoflavum</i>	read: <i>E. roseoflavum</i>
p.506, line 23                    for: <i>Symphaster ximeniae</i> ....., p. 2	read: <i>Symphaster ximeniae</i> ....., p. 220
p. 517, line 14                for: (See pp. 515–515, this volume)	read: (See pp. 514–515, this volume)

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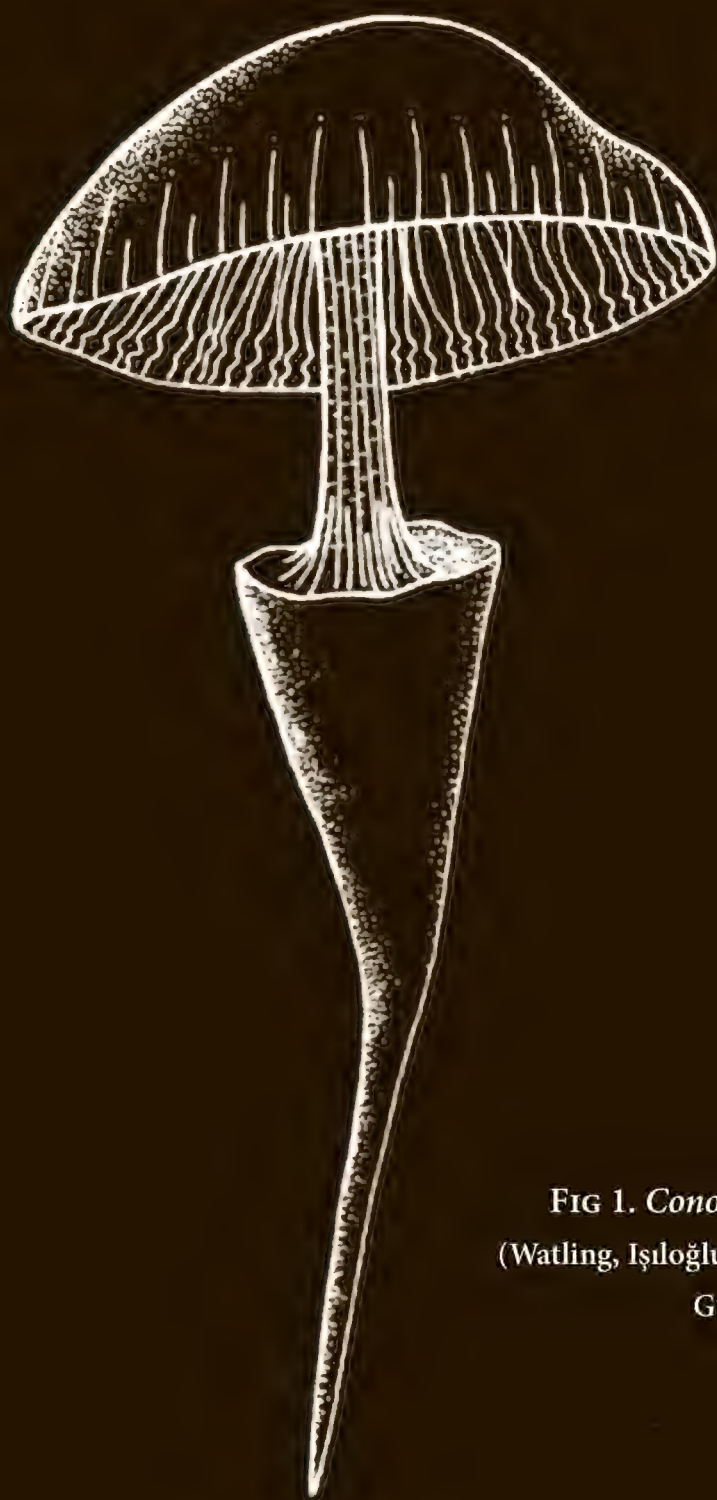


FIG 1. *Conocybe volviradicata* sp. nov.  
(Watling, Işıloğlu & Baş Sermenli— FIG. 1, p. 147)  
GÜLNUR EKŞİ, artist

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## MYCOTAXON

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***Caloplaca tianshanensis* (lichen-forming Ascomycota),  
a new species of subgenus *Pyrenodesmia* from China**HURNISA XAHIDIN<sup>1,2</sup>, ABDULLA ABBAS<sup>1</sup> & JIANG-CHUN WEI<sup>3\*</sup>*Hurnisa\_xju@sina.com & weijc2004@126.com or weijc@im.ac.cn*<sup>1</sup>*College of Life Science and Technology*<sup>2</sup>*College of Resource and Environment Sciences**Xinjiang University, Urumqi 830046, P. R. China*<sup>3</sup>*Key Laboratory of Systematic Mycology & Lichenology,**Institute of Microbiology, Chinese Academy of Sciences**1-3 West Beichen Road, Chaoyang District, Beijing 100101, P. R. China*

**Abstract** — *Caloplaca tianshanensis* is described as a species new to science. It has a crustose and areolate thallus of yellowish-brown color with conspicuous cracks, bearing dark brown to black apothecia. An analysis of ITS sequences supports the affinity of the new species to subgenus *Pyrenodesmia*.

**Key words** — *Teloschistaceae*, peltate areoles, zeorine, isthmus

**Introduction**

As presently circumscribed, the subgenus *Pyrenodesmia* (A. Massal.) Boistel of the lichen-forming genus *Caloplaca* Th. Fr. (*Teloschistaceae*) contains lichens characterized by brown or black apothecia, an epihymenium that is usually K– or K+ violaceous, and a thallus that is not yellow, orange or red unlike most other *Caloplaca* spp., and lacks the K+ red reaction of the parietin complex (Tretiach & Muggia 2006).

Forty-two species of the genus *Caloplaca* were reported from China (Wei 1991). Among them 9 species belong to the subgenus *Pyrenodesmia*: *C. chrysophora* Zahlbr., *C. cupreorufa* Zahlbr. and *C. cervina* Zahlbr. from Sichuan (Zahlbruckner 1930, 1932), *C. giraldii* Jatta from Shaanxi (Jatta 1902) and Sichuan (Zahlbruckner 1930, 1931), *C. ochrotropa* Zahlbr. from Yunnan (Zahlbruckner 1930, 1932), *C. plumbeoolivacea* H. Magn., *C. circumalbata* (Delile) Wunder from Inner Mongolia (Magnusson 1944, as *C. aegyptiaca* (Müll.

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\*corresponding author

TABLE 1. Lichen species and sequences used to generate the phylogenetic tree.

SPECIES	GENBANK #
<i>Caloplaca albopruinosa</i> (Arnold) H.Olivier	EF093577
	EF093578
<i>C. albopustulata</i> Khods. & S. Y. Kondr.	EU192150
<i>C. alociza</i> (A. Massal.) Mig.	EF090933
	EF090936
<i>C. badioreagens</i> Tretiach & Muggia	EF081039
	EF081040
<i>C. cerina</i> (Ehrh.Ex Hedw.) Th.Fr.	AF353958
<i>C. chalybaea</i> (Fr.) Müll.Arg.	AY313970
	AY313971
<i>C. chlorina</i> (Flot.) Sandst.	AF353959
<i>C. concreticola</i> Vondrák & Khodos.	EU192153
	EU192152
<i>C. cretensis</i> (Zahlbr.) Wunder	EF093579
<i>C. erodens</i> Tretiach <i>et al.</i>	EF090922
	EF090921
<i>C. obscurella</i> (J. Lahm) Th.Fr.	AY313976
	AY313977
<i>C. peliophylla</i> (Tuck.) Zahlbr.	AY313965
<i>C. tianshanensis</i> Xahidin, A. Abbas & J.C. Wei <sup>a</sup>	GU552277
<i>C. transcaspica</i> .	EU192156
<i>C. variabilis</i> (Pers.) Müll. Arg.	EF090926
	EF090925

Arg.) Stnr; Wunder 1974), *C. transcaspica* (Nyl.) Zahlbr. from Inner Mongolia (Magnusson 1944, as *C. paulsenii*), Gansu, Qinghai (Magnusson 1940, as *C. paulsenii*) and Xinjiang (Poelt & Hinteregger 1993)., and *C. alociza* (Massal.) Mig. from Jiangsu (Wu & Xiang 1981, as *C. agardhiana* (Flot.) Flag., 1981).

During a study of the lichen genus *Caloplaca* in China numerous samples were collected by the first two authors from the Xinjiang region. Some specimens belonging to *Pyrenodesmia* attracted our special attention and were examined in detail for morphology, anatomy, chemistry and molecular systematics. As a result, one of them, *C. tianshanensis*, is described here as new to science.

Material and methods

Material

The lichen material examined for morphology, anatomy, chemistry and molecular analyses was collected from Miaoergou on Mt. Nan-shan in the Tianshan mountain chain, Xinjiang region, in 2009.



## Morphological observations

Observations and photographs were made with a dissecting microscope (Leica MZ 12), a Zeiss Axioplan compound microscope and an Axiocam digital camera with associated software. Squash mounts and hand sections were routinely examined using tap water as the mounting medium. Lichen substances were detected by TLC and MCT (Culberson & Kristinsson 1970, Culberson 1972, Orange et al. 2001).

## DNA extraction, amplification, and sequencing

The dried apothecia first were checked under the dissecting microscope for well-developed fruit bodies to avoid contamination of other organisms.

Total DNA was extracted from dry apothecia following the rapid one-tube genomic DNA extraction (Steiner et al. 1995) with modifications: seven dried and cleaned apothecia were transferred directly into a 2 ml Eppendorf tube. The material was grinded with a pestle in liquid nitrogen until a fine powder was obtained. Then 150 µl TE solution was added into the tube and stirred for 2 min. until the powder was well-distributed, and immediately stored at –20°C.

Primers for PCR of the nuclear ribosomal ITS region ITS1F (Gardes & Bruns 1993) and ITS4 (White et al. 1990) were used.

The phylogenetic tree was constructed with a Bayesian approach based on the nuclear ribosomal ITS sequence data of the new species and sequences of species from the same subgenus retrieved from GenBank (TABLE 1).

## Taxonomy

***Caloplaca tianshanensis*** Xahidin, A. Abbas & J.C. Wei, sp. nov. (FIGS 1, 2)

MYCOBANK MB 518332

*Species nova similis C. peliophyllae a qua thallo flavido-brunneo areolato cum rimis conspicuis et areolis peltatis, stipitatis in centro thalli, discis apotheciorum atris raro atrobrunneis, substantias lichenium ignotas continente differt.*

TYPE: China, Xinjiang, Mt. Nan-shan in Tianshan mountain chain, Miaoergou, on limestone, alt. 1280 m, April 10, 2009, A. Abbas & H. Xahidin 20090001 (**holotype** in XJU, **isotype** in HMAS–L).

ETYMOLOGY: The specific epithet refers to the type locality.

THALLUS crustose, 2–11 cm in diam., consisting of numerous peltate areoles of 0.7–3 mm wide and 0.4–0.6 mm thick, much thicker in central part of the thallus, yellowish brown, flat, separated by conspicuous cracks (FIG. 1a, b), with a whitish gray to light gray and very thin prothallus.

Upper cortex well developed, paraplectenchymatous, 50–175 µm thick; algal layer discontinuous (FIG. 1c).

ASCOMATA apothecia, orbicular to irregular in shape, immersed or somewhat prominent, 0.8–1 mm in diam., numerous, usually 1 per areole, sometimes 2 or occasionally more than 2, zeorine, with both a proper and a thalline margin; thalline margin raised and proper margin not visible when younger;

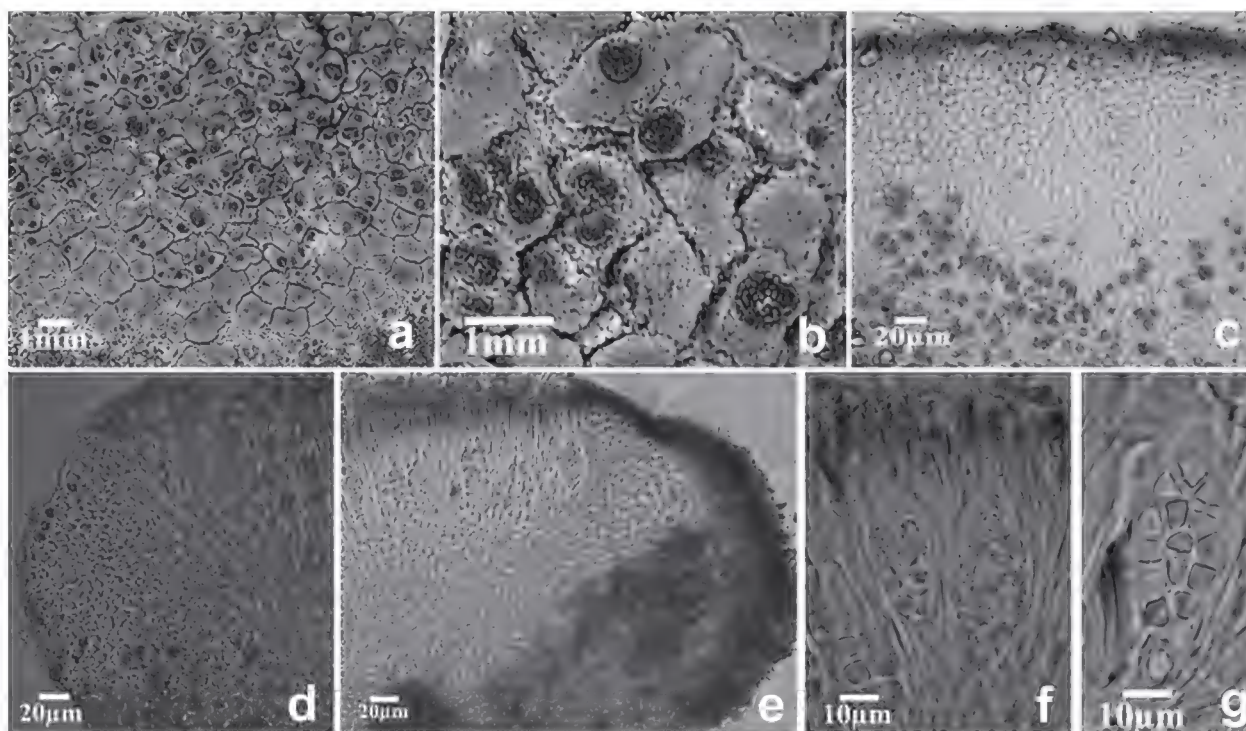


FIG.1. *Caloplaca tianshanensis*: a, b. habit; c. cross section of a peltate areole of the thallus showing the well-developed paraplectenchyma in the upper cortex; d. cross section of an apothecium showing the well-developed paraplectenchyma in the proper exciple; e. cross section of an apothecium showing the double or zeorine margin, with both thalline and proper exciple; f. cross section of the hymenium showing asci containing spores and paraphyses with beaded apices consisting of 2–5 swollen terminal cells; g. an ascus containing 8 spores with thin septa.

proper margin raised and prominent, and thalline margin lower when mature (FIG. 1e); disc dark brown to black, concave, shiny, without or with thin whitish pruina (FIG. 1a, b); hymenium 75–115  $\mu\text{m}$  thick; paraphyses septate, simple, with beaded apices consisting of 2–5 swollen cells (FIG. 1f); asci 44–62  $\times$  12–26  $\mu\text{m}$ , 8-spored; spores broadly ellipsoid, polarilocular, 12–18  $\times$  5–9  $\mu\text{m}$  (FIG. 1f, g); proper exciple paraplectenchymatous (FIG. 1d); hypothecium with gray crystals, 55–90  $\mu\text{m}$  thick.

CONIDIOMATA not seen.

CHEMISTRY: upper cortex K–, C–, epihymenium K–; two unknown substances were detected by TLC: one gives a spot in  $R_f$  class 5–6 by solvent systems A, B and G, and in  $R_f$  class 6 by solvent system C, grey-brown after charring; the other gives a spot in  $R_f$  class 5 by solvent systems A and G, in  $R_f$  class 2 by B, and in  $R_f$  class 2–3 by C, green after charring.

REMARKS: The new species is similar to *C. peliophylla* in its yellowish brown thallus, but different by the areolate thallus, dark brown to black apothecium discs, the presence of two unknown lichen substances, and the Asian distribution. The latter species differs in its subsquamose thallus with shiny brown apothecia, an American distribution and the absence of lichen substances (Wetmore 1994). In addition, the new species is similar to *C. transcaspica* in its crustose and

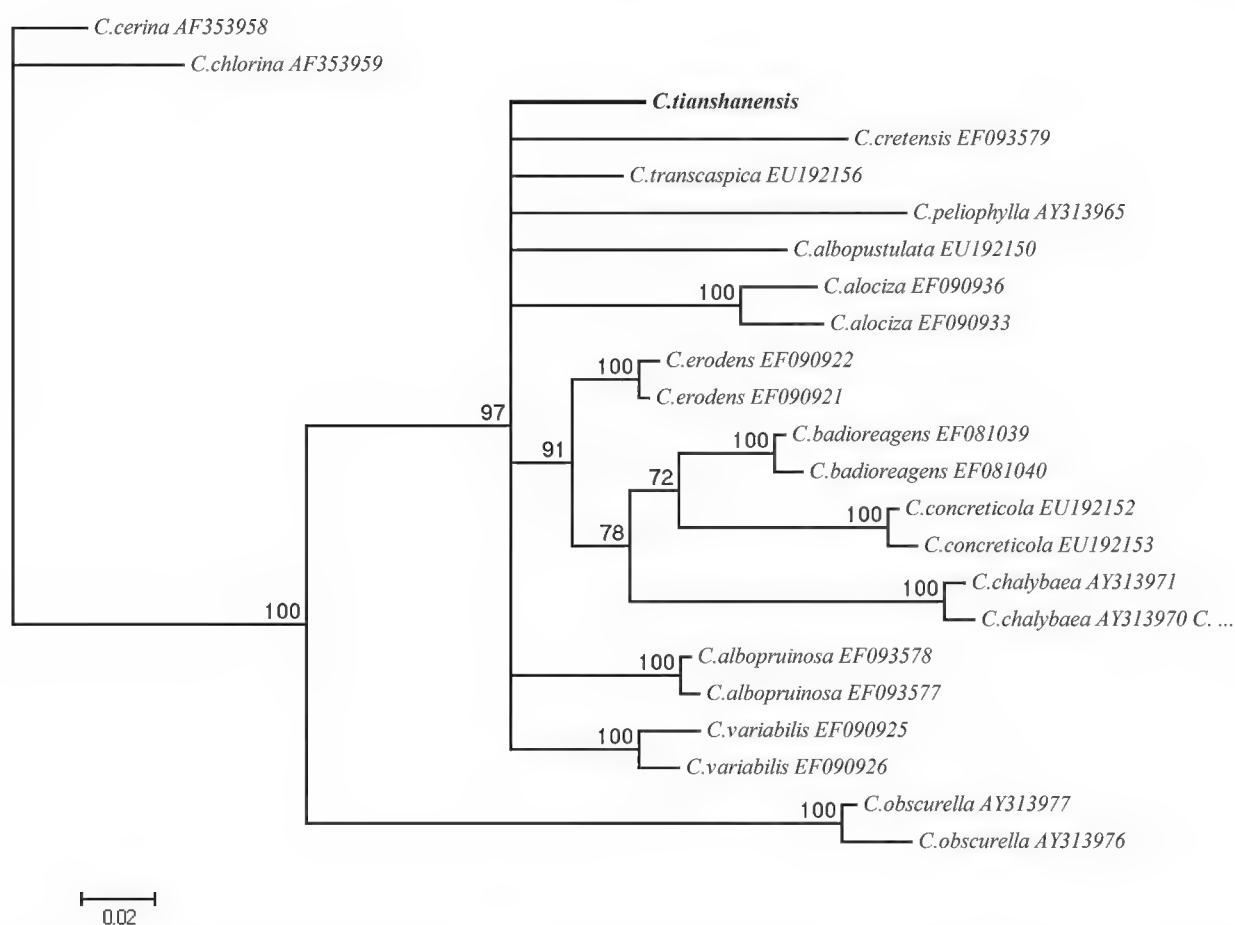


FIG. 2. Consensus tree generated by Bayesian analysis based on ITS region rDNA sequence data. *Caloplaca tianshanensis* groups with species of subgenus *Pyrenodesmia*. Bootstrap support values from 1000 replicates higher than 50% are reported at the nodes. *C. cerina* and *C. chlorina* from subgenus *Caloplaca* were selected as outgroup.

areolate thallus, but differs by its yellowish brown color, dark brown to black discs, smaller ascospores, wider isthmus in cells, and 2–5 swelling terminal cells of the paraphyses.

The ITS sequence of *C. tianshanensis* grouped with those of other 12 related species was retrieved from GenBank as a group belonging to the subgenus *Pyrenodesmia* with 100% bootstrap support. The ITS sequence of the new species *C. tianshanensis* form a distinct clade among the other 11 well recognized related species, such as *C. cretensis*, *C. transcaspica*, *C. peliophylla*, *C. albopustulata*, etc., with 97% bootstrap support. These results show that *C. tianshanensis* is clearly distinct from the above-mentioned well-recognized species (FIG. 2).

Acknowledgments

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## MYCOTAXON

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**A new species of *Physarum* (Myxomycetes)  
from a boreal pine forest in Thuringia (Germany)**

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**Abstract** – A new species of plasmodial slime mold, *Physarum parvicalcareum*, recorded from flowering stalks, leaves and stems of the common heather (*Calluna vulgaris*) that commonly inhabits boreal pine forests in Thuringia (Germany), is described based on morphological features of the capillitium and spores. Phylogenetic trees, reconstructed using data from elongation factor-1 alpha and small subunit ribosomal RNA gene sequence analyses, corroborate the taxonomic status of this new species. In addition, these results support the congruence of morphological and molecular data in this group of eukaryotic microorganisms.

**Keywords** – molecular phylogenetics, myxomycetes, species concepts

**Introduction**

Plasmodial slime molds (*Mycetozoa*, commonly referred to as myxomycetes) have been regarded as either “plant-like animals” or “animal-like plants,” depending on whether a zoologist or a botanist investigated them. In the 19th century, taxonomists classified the myxomycetes as either fungi or protozoans (Martin & Alexopoulos 1969). However, a detailed analysis of DNA sequence data has recently shown beyond any doubt that these inhabitants of soil and other habitats containing moist, decomposing organic matter comprise a sister taxon to the *Amoebozoa* and hence are members of the Kingdom *Protocista* (Pawlowski & Burki 2009, Hoppe & Kutschera 2010).

*Physarum* is the most widely known genus among the myxomycetes, due to the fact that the species *P. polycephalum* serves as a model organism for cell research.

Several years ago, Müller (2007) collected an unidentifiable myxomycete in boreal forests in the Federal State of Thuringia (Germany). Based on morphological, ultrastructural, and molecular data, we herein describe this taxon as a new species of the genus *Physarum*.

**NOTE** —MYCOTAXON PREPARED THIS PDF WITH COLOR PLATES FOR THE AUTHOR. THE ORIGINAL PRINT VERSION WAS PUBLISHED WITH HALFTONE (GRAYSCALE) PLATES.



## Materials and methods

During field trips in 2005 and two subsequent years to the boreal pine (*Pinus sylvestris* L.) forests in the Federal State of Thuringia (close to the towns of Rudolstadt and Mörla in eastern Germany, Central Europe), samples were collected from the leaves, stems, and flowering stalks of the common heather, *Calluna vulgaris* (L.) Hull. This cut plant material was analyzed in the laboratory, using a stereo light microscope (Photomikroskop III, Carl Zeiss, Germany) and a scanning electron microscope (REM, S-4000, Hitachi, Japan) as described by Hoppe & Kutschera (2010). Sample preparations and photographic documentation of the results were carried out as described in the reference cited above. Collections are conserved in Botanische Staatssammlung München (M) and the private collections of T. Hoppe (Germany), H. Müller (Germany), M. Meyer (France), and W. Nowotny (Austria).

Extraction of total deoxynucleic acids (DNA), DNA-amplification via polymerase chain reaction (PCR), and phylogenetic analyses were performed as described in Hoppe & Kutschera (2010). In brief, fruiting bodies were mechanically crushed, after which the homogenized samples were first treated using a FastRNA Pro Red Kit (Solon, Colorado, USA) and then incubated for 90 sec. with the Fast Prep-System FP120 (MP Biomedical) (Costa et al. 2004). In the next step, the samples were incubated for 24 h in a solution of lysozyme (5%, 35°C) and thereafter for 24 to 48 h in proteinase K (5%, 55°C) (Roth, Karlsruhe, Germany). DNA-purifications were performed using a QIAamp DNA Mini Kit (Quiagen, Hilden, Germany). The purified DNA samples were amplified with primers designed for specific elongation factor-1 $\alpha$  gene sequences (Hoppe & Kutschera 2010) and primers for the small subunit of a ribosomal RNA gene (Kamono & Fukui 2006). The products were purified using NucleoSpin Extract II (Machery-Nagel, Germany) and sequenced. Phylogenetic trees, based on maximum parsimony analyses, were reconstructed as described by Hoppe & Kutschera (2010).

## Taxonomy

***Physarum parvicalcareum*** Thom. Hoppe, Holg. Müll. & Kutschera, **sp. nov.**

MYCOBANK MB 516617; NCBI (GENBANK) FJ 558512 AND GU 289193

FIGS. 1–4

*Sporocarpia sessilia, singula vel gregaria vel seria, globosa vel semiglobosa vel brevia plasmodiocarpia, violaceus usque ad aeneus iridescens, (0.3–)0.6–0.8 mm in diametro, usque ad 2 mm longae. Hypothallus membranaceus, translucidus. Peridium simplex, calce non incrustatum, violaceus usque ad aeneus, nonnullum clarum zonatum iridescens, lucem orientem versus visae incoloratum, dehiscencia irregularis. Capillitium reticulatum, album vel alutaceum, lucem orientem versus visae incolor, capilloides vel fasciatum, (1–)2–5(–7)  $\mu$ m in diametro, cum nodis calcareis parvis. Columella vel Pseudocolumella nulla. Sporae frequentes brunneus, lucem orientem versus visae cineraceo-brunneae vel brunneus, globosae, dense cum obscurus, irregulariter verrucosae, 10–11(–12)  $\mu$ m in diametro. Plasmodium ignotum.*

TYPE SPECIMENS: Germany, close to Mörla, 50.43°N 11.20°E, on stems, green leaves and flowering stalks of *Calluna vulgaris* in a pine forest, 10 Oct. 2005, Holger Müller. (**Holotype**: Botanische Staatssammlung München (Germany), M 0151322; **Isotype**: private collection of H. Müller (Germany), Müll. 2238).

ETYMOLOGY: from the Latin *parvus* = small; *calcareus* = calcareous.

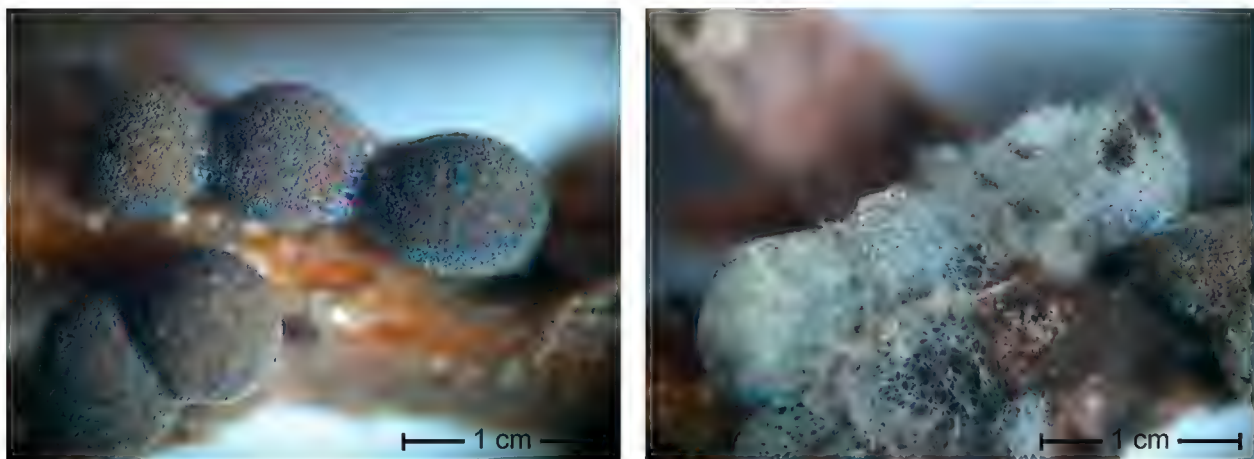


FIG. 1. Photographs of the sporocarps of *Physarum parvicalcareum* attached to a flowering stalk of *Calluna vulgaris*. A- Sporocarps with peridium still intact and spores present. B- Sporocarps after the release of the spores.

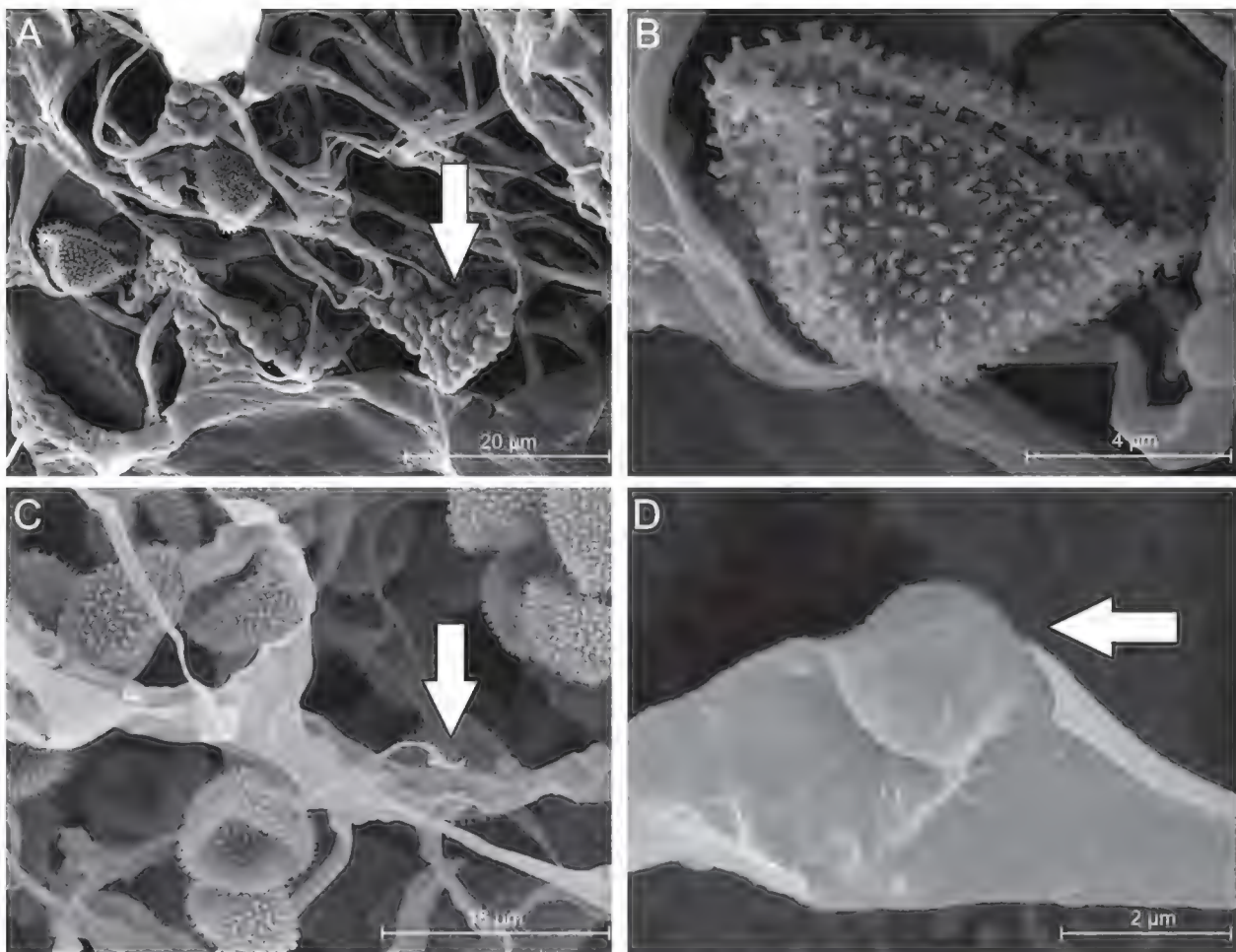


FIG. 2. Scanning electron micrographs of capillitium and spores of *Physarum parvicalcareum*. A- Capillitium with calcareous deposits (arrows). B- Single spore. C- Spores with capillitium that is free of deposits. D- Isolated portion of smooth capillitium with grain (calcareous deposit).

Sporocarps sessile, single, in groups or in lines, globose or sub-globose or short plasmodiocarps, violet to bronze, iridescence in white light, (0.3–)0.6–0.8 mm in diameter, up to 2 mm long (FIG. 1A,B). Hypothallus membranous, transparent, continuous within a group of fruiting bodies. Peridium single,

TABLE 1: Morphological features of *Physarum parvicalcareum*, *P. nudum*, and *P. cinereum*, based on newly collected and herbarium materials

CHARACTER	<i>P. parvicalcareum</i>	<i>P. nudum</i>	<i>P. cinereum</i>
Mature fruiting body	sporocarp or plasmodiocarp	sporocarp or plasmodiocarp	sporocarp or plasmodiocarp
– colour	violet to bronze	white to faint violet	white
– diameter (mm)	0.6–0.8	0.4–1.0	0.3–0.6
– length (mm)	up to 2.0	up to 1.2	up to 0.8
Capillitium surface	smooth or rough	rugged	rugged
– pili length (nm)	up to 300	up to 300	up to 250
Calcareous deposits within fruiting bodies	greatly reduced or absent	present	present

limeless, violet to bronze, some individuals conspicuously iridescent, colourless in transmitted light, dehiscence irregular. Columella or pseudo-columella absent. Capillitium consists of a three-dimensional net with small meshes, white to pale-yellow, colourless in transmitted light, filamentous, with few swellings, covered with small lime granules (FIG. 2A–D), sometimes with a band-like widened appearance, (1–)2–5(–7) µm in diameter. Spores dark-brown, grey-brown or dark-brown in transmitted light, globose, 10–11(–12) µm in diam., densely covered with dark, coarse, more or less irregular warts. Plasmodium not observed.

ECOLOGY AND HABITAT – Living stems, leaves, and flowering stalks of *Calluna vulgaris*; no fruiting bodies were found on nearby plants in the same area.

EXPANDED DESCRIPTION – A phylogenetic analysis, based on novel elongation factor-1 alpha gene sequences supplemented by published data, is depicted in FIG. 3. In addition, a partial (123 bp) sequence of the small subunit of a ribosomal RNA gene was investigated (GenBank-Numbers provided above the lines in FIGS. 3, 4) and aligned with morphologically similar species. These data show that *Physarum parvicalcareum* is closely related to the species *P. cinereum* and *P. nudum*, but differs from these taxa in several morphological features (TABLE 1). Hence, our evolutionary trees (FIGS. 3, 4), in tandem with our morphological data (FIGS. 1, 2) document *P. parvicalcareum* as a new species and not a morphological variant (variety) of *P. nudum* or one of the other taxa that were analyzed as part of the present study.

ADDITIONAL SPECIMENS EXAMINED: GERMANY, close to Mörla, 50.43°N 11.20°E, on stems, green leaves and flowering stalks of *Calluna vulgaris* in a pine forest, 15 Oct. 2005, Holger Müller (Distributed among private collections of H. Müller (Germany: Müll. 2632); M. Meyer (France: 29759, 29760, 30078, 30077); T. Hoppe (Germany: Myx 90); and W. Nowotny (Austria: Now. 13507).

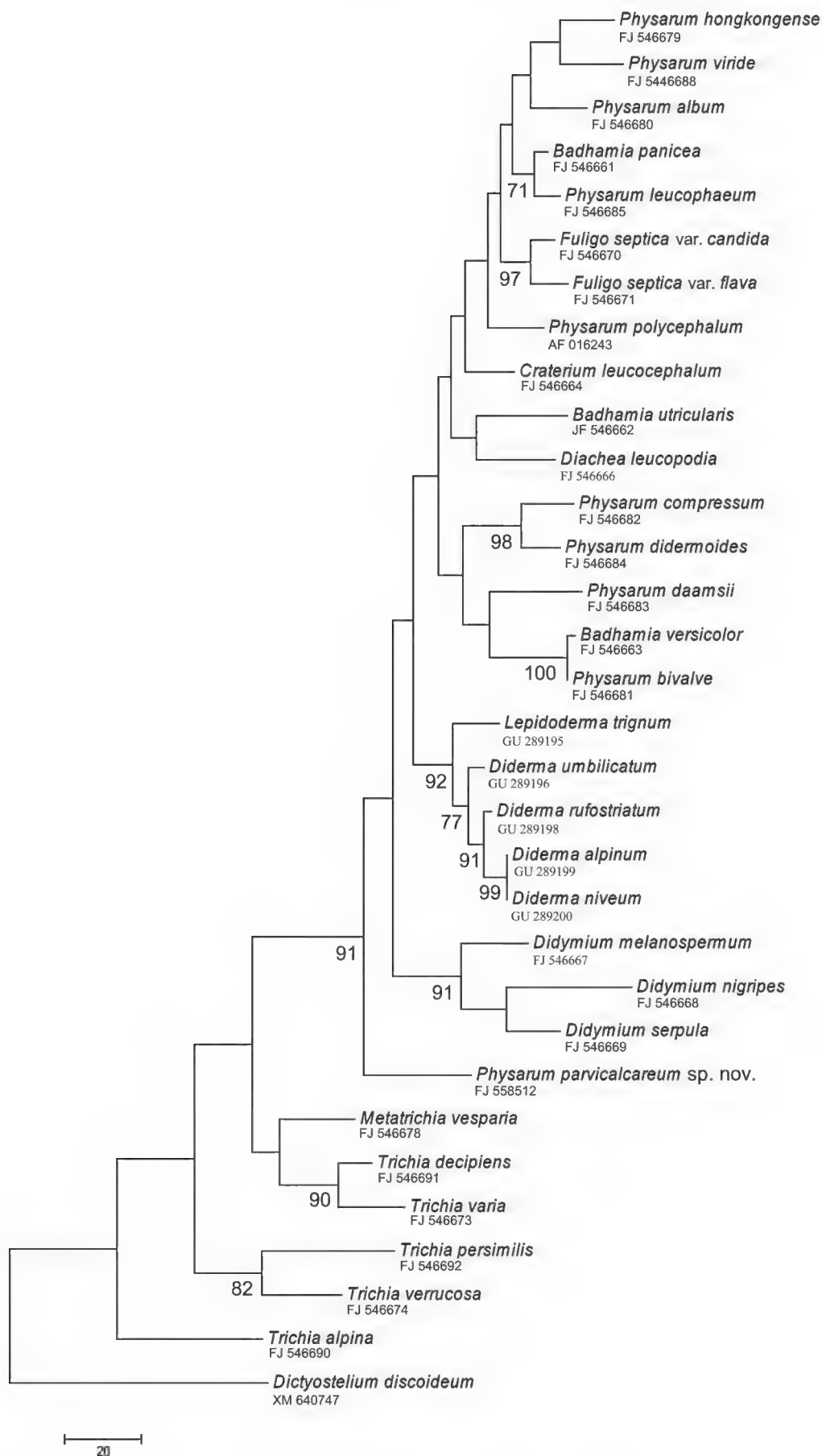


FIG. 3. Phylogenetic tree based on elongation factor-1 alpha gene sequences of 31 myxomycetes, with *Dictyostelium discoideum* as outgroup. The bootstrap values of this maximum parsimony analysis and the GenBank accession numbers are included.



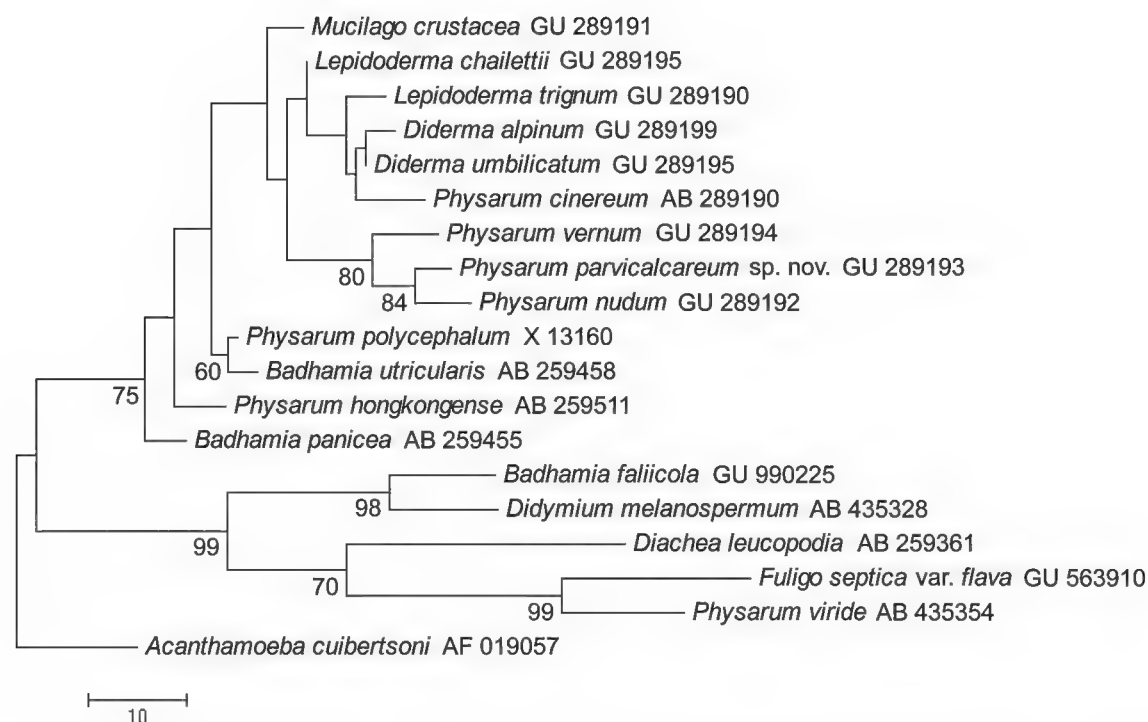


FIG. 4. Phylogenetic tree based on a fragment (123 bp) of the small subunit of a ribosomal RNA gene of 18 morphologically distinct species, with *Acanthamoeba cuibertsoni* as outgroup. The bootstrap values of this maximum parsimony analysis and the GenBank accession numbers are included.

COMMENTS – As pointed out by Neubert et al. (1993, 1995, 2000) and Clark (2000, 2004), myxomycete taxonomy is currently based on the classical morphological species concept. Hence, myxomycetologists have described numerous taxa (“species”) that were found in a single habitat or a restricted area, often in low numbers, or even based on a single individual (Lado 2001).

As a result of his survey of recent biosystematic studies in the myxomycetes, Clark (2004) suggested that many commonly accepted morphospecies might, in reality, be species complexes. Moreover, the author suggested that many of the accepted morphospecies might represent morphological variants of one and the same “true” species and therefore should be assigned to the same taxon. In conclusion, Clark (2004) recommended that new species be described only on the basis of numerous collected specimens from different localities so that the problems outlined above could be circumvented.

In this report we describe a new species of myxomycete that was assigned to the genus *Physarum*. The question to be discussed here is whether or not we have met the standards proposed by Clark (2004). In other words, is *P. parvicalcareum* (FIG. 1, 2) only a “variant” of a closely related taxon or does it in fact represent a truly new species?

Our arguments in support of the second conclusion can be summarized as follows. First, the requirement that a new species should be based on an



extensive collection of individuals made on different occasions and (if possible) localities has been met. As documented in a previous report (Müller 2007), numerous samples were collected on several different occasions in Thuringia, and over the past year we found additional specimens of *P. parvicalcareum* in forests in close proximity to the type locality described above (unpublished observations). Second, there is sufficient morphological evidence to separate our new species from all other taxa assigned to the genus *Physarum*, particularly the most closely related species (TABLE 1). Finally, we analyzed the phylogenetic relationships among 30 myxomycete species within the genera *Trichia*, *Hemitrichia*, and *Metatrichia* in the order *Trichiales* and within the genera *Badhamia*, *Fuligo*, *Craterium*, *Diachea*, *Didymium*, and *Physarum* in the order *Physarales*. These analyses were based on elongation factor-1 alpha gene sequences. In addition, 14 relevant species of the *Physarales* belonging to the genera *Badhamia*, *Diachea*, *Didymium*, *Lepidoderma*, *Physarum* (8 different species), and *Mucilago*, based on a partial sequence of the small subunit of a ribosomal RNA gene, were also investigated. Our quantitative maximum parsimony analyses (FIGS. 3, 4) led to the conclusion that *P. parvicalcareum* represents a distinctly new species and not a “morphological variant” of another taxon assigned to the genus *Physarum*.

In summary, our results document that on the above-ground portions (leaves, stems, and flowering stalks) of the common heather (*Calluna vulgaris*) there occurs a myxomycete species that is described here as *P. parvicalcareum*. However, we do not yet know whether or not this new *Physarum* species inhabits its host organism as a commensal or an endophytic parasite (Stephenson & Studlar 1985). It should be noted that we are currently unaware of any other plant species in the boreal pine forest where our new myxomycete was discovered that is inhabited (or infected) by *P. parvicalcareum*. However, more fieldwork is required to further elucidate the entire habitat of this new plant-associated species of the genus *Physarum*.

### Acknowledgements

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## MYCOTAXON

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***Muscodor cinnamomi*,  
a new endophytic species from *Cinnamomum bejolghota***

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**Abstract** — *Muscodor cinnamomi* is described as a new species, endophytic within leaf tissues of *Cinnamomum bejolghota* (*Lauraceae*) in Doi Suthep-Pui National Park, Northern Thailand. Molecular analysis indicated differences from the five previously described *Muscodor* spp. Volatile organic compounds analysis showed that *M. cinnamomi* produced azulene (differentiating it from *M. crispans*) but did not produce naphthalene (differentiating it from *M. albus*, *M. roseus*, and *M. vitigenus*).

**Key words** — sterile ascomycete, cinnamon, endophytes, volatile compounds

## Introduction

Plants are reservoirs of untold numbers of endophytic organisms (Bacon & White 2000). By definition, these microorganisms (mostly fungi and bacteria) reside in the tissues beneath the epidermal cell layer and cause no apparent harm to the host (Azevedo et al. 2000, Hyde & Soyong 2008). Endophytes from rainforest and medicinal plants have been studied for their volatile antibiotic and other medicinal characteristics (Strobel et al. 2003, Huang et al. 2008, 2009, Mitchell et al. 2008, Tejesvi et al. 2009, Aly et al. 2010). Five endophytes characterized by sterile mycelium that have recently been described as novel fungi are *Muscodor albus* isolated from *Cinnamomum zeylanicum* (*Lauraceae*) in Honduras (Worapong et al. 2001), *M. roseus* from *Grevillea pteridifolia* (*Proteaceae*) in the Northern Territory of Australia (Worapong et al. 2002), *M. vitigenus* from *Paullinia paullinioides* (*Sapindaceae*) in Lake Sandoval (Daisy et al. 2002), *M. crispans* from *Ananas ananassoides* (*Bromeliaceae*) in the Bolivian Amazon (Mitchell et al. 2008), and *M. yucatanensis* from *Bursera*

*simaruba* (*Burseraceae*) in the Northeastern Yucatan Peninsula of Mexico (González et al. 2009). All *Muscodor* species grow slowly, have felt-like mycelia, and produce a distinctive odor. Gas chromatography and mass spectrometry (GC/MS) can be used to identify *Muscodor* species based on differences in the volatile compounds that they produce (Strobel et al. 2001).

In the present study an endophyte (CMU-Cib 461) was recovered from leaf tissue of a wild cinnamon tree (*Cinnamomum bejolghota*) growing in Doi Suthep-Pui National Park, Thailand. The strain produced a mixture of volatile compounds including propanoic acid and alcohol; these have antagonistic activities and can be used to identify the particular *Muscodor* species. CMU-Cib 461 possesses cultural, chemical, and molecular characteristics that differ from *M. albus*, *M. crispans*, *M. roseus*, *M. vitigenus*, and *M. yucatanensis*. We conclude that CMU-Cib 461, based on its unique features, represents a new species of *Muscodor*, for which we propose the name *Muscodor cinnamomi*.

## Materials and methods

### Fungal isolation

Ten healthy leaves of *Cinnamomum bejolghota* were collected from plants growing in Doi Suthep-Pui National Park, Northern Thailand (alt. 950 m) during May 2008. Totally, 250 tissue squares (5 × 5 mm) were cut from the leaf samples. All leaf tissues squares were surface sterilized in 75% ethanol for 30 s, 2% sodium hypochlorite for 3 min and 95% ethanol for 30 s under a laminar flow hood (Nuangmek et al. 2008). The sterilized samples were placed in Petri dishes containing 2% malt extract agar, 0.05% streptomycin sulfate and 0.03% rose bengal (Bussaban et al. 2001). Petri dishes were sealed with Parafilm and incubated at room temperature (25±2°C) for one week. The fungi growing out from the samples were aseptically transferred to two culture media, potato dextrose agar (PDA) and malt extract agar (MA); pure isolates were maintained in corn meal agar (CMA) slants. Various methods were tried to stimulate spore production (Guo et al. 1998).

### Scanning electron microscopy

Scanning electron microscopy was performed on isolate CMU-Cib 461 following procedures described by Castillo et al. (2005). A piece of agar with fungus was placed in a filter paper packet and then placed in 2% glutaraldehyde vapor, a wetting agent, and aspirated over night. Samples were then dehydrated in an ethanol series (15 mins at 5, 10, 15, 20, 40, 50, 70, 80, 95 and 100%) and in an acetone series (10 mins at 10, 15, 20, 40, 50, 70, 80, 95 and 100%). The fungal material was critically point dried, gold sputter coated, and images observed under a JEOL JSM-5910LV SEM using a high vacuum mode.

### Qualitative analysis of CMU-Cib 461 volatiles

CMU-Cib 461 was grown in 5 ml Agilent® clear glass vials containing PDA for 10 days at room temperature (25±2°C). Volatile compounds produced by the fungus were analyzed on an automatic Agilent Technologies GC 7890 gas chromatograph column



containing a HP-5MS 30 m × 0.25 mm I.D. × 0.25 µm. The column was temperature programmed as follows: 32°C for 2 min followed to 220°C at a rate of 5°C/min. The carrier gas was ultra high purity helium released at a rate of 1.5 mL/min. Prior to trapping the volatiles, the fiber was conditioned at 250°C for 39.6 min under a flow of helium gas. The gas chromatograph was interfaced to a MSD 5973 (EI) mass selective detector (mass spectrometer) operating at unit resolution. Acquisition and processing data were performed on the MSD 5973 (EI) software system. Initial identification of the volatile compounds produced by CMU-Cib 461 was made through library comparison using the NIST database, and compared with the original isolates, *M. albus* strain 620 (Strobel 2006) and strain E-6 (Strobel et al. 2007).

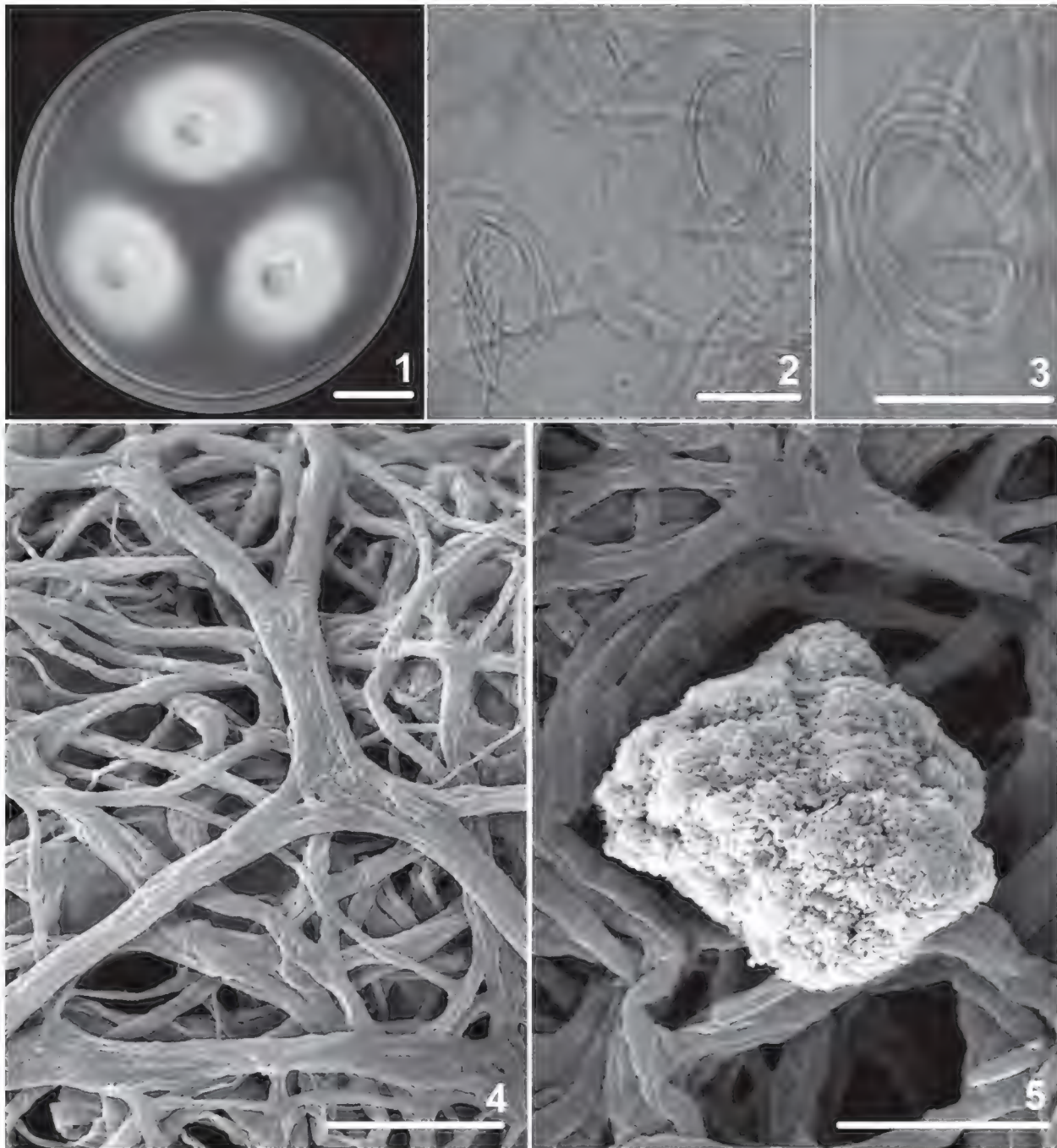
### **Fungal cultures and DNA extraction**

Genomic DNA was extracted by a modified SDS-CTAB method (Bussaban et al. 2005). Strain CMU-Cib 461, isolated from *C. bejolghota* leaves, was subcultured onto PDA and incubated for 10 days. Mycelium was harvested, freeze dried, and ground into a fine powder with a pestle and mortar. About 15 mg of powdered mycelium was suspended in 1 mL of ice-cold lysis buffer (150 mM NaCl, 50 mM EDTA, 10 mM Tris-HCl, pH 7.4, 20 mg/mL proteinase K), transferred into 1.5 mL Eppendorf tube and kept at 4°C to prevent endonuclease activity during rehydration of the sample. SDS was added to a final concentration of 2%, vortexed and incubated 30 min at 65°C. After centrifugation for 15 min at 14,000 rpm, the supernatant was transferred to a new sterile 1.5 mL Eppendorf tube. The volume of supernatant was measured and the NaCl concentration was adjusted to 1.4 M, and one-tenth volume of 10% CTAB buffer (10% CTAB, 500 mM Tris-HCl, 100 mM EDTA, pH 8.0) was added. The solution was thoroughly mixed and incubated for 10 min at 65°C. After cooling for 2 min at 15°C, an equal volume of chloroform: isoamyl alcohol (24:1 v/v) was added, thoroughly mixed and the tube was centrifuged 15 min at 14,000 rpm. The extraction was repeated until the interface was clear. The supernatant was removed to a new Eppendorf tube, containing 2 volumes of cold 100% ethanol. After DNA precipitation, the pellet was centrifuged for 15 min at 14,000 rpm and 4°C. The pellet was washed with 70% ethanol and dried at room temperature. It was resuspended in 100 µL of 0.002% RNase (5 mg/mL) in TE buffer and incubated for 1 h at 37°C. The suspension was stored at -20°C pending use for PCR amplification.

### **Fungal ITS regions sequencing and phylogenetic analysis**

The internal transcribed spacer (ITS) regions 1 and 2, including 5.8S rDNA were separately amplified in a 25 µL reaction on a GeneAmp 9700 thermal cycler (Applied Biosystems) under these reaction conditions: 1 µL of template DNA extraction, 0.2 mM dNTP, 0.2 µL of FastTaq (Applied Biosystems), 0.2 mM each of primers, 2.5 µL of the supplied 103 PCR buffer with MgCl<sub>2</sub>, and sterile water to bring the volume to 25 µL. The ITS regions were amplified by using ITS4 and ITS5 primers. Amplification of ITS regions was for 30 cycles (initial denaturation at 95°C for 2 min, denaturation at 95°C for 30 s, annealing at 50°C for 30 s, and extension at 72°C for 1 min, with a final extension at 72°C for 10 min). PCR products were analyzed by electrophoresis in 1% agarose gels in TAE buffer (20 mM Tris-Acetate, 1 mM EDTA, pH 8.0) and viewed by staining with ethidium bromide. PCR products were purified using PCR clean up Gel extraction





FIGS. 1–5. *Muscodor cinnamomi* 1. A culture of *Muscodor cinnamomi* CMU-Cib 461 growing on PDA, bar = 2 cm. 2–3. Light microscope micrographs of coiling formation of fungal hyphae, bars = 5  $\mu$ m. 4–5. Scanning electron micrographs. 4. Hyphal cells from the colony edge showing fused, rope-like hyphal cells, bar = 10  $\mu$ m. 5. Fused hyphal cells and a cauliflower-like structure, bar = 5  $\mu$ m.

NucleoSpin® Extract II purification Kit (Macherey-Nagel, Germany) following the manufacturer's protocol. The purified PCR products were directly sequenced. Sequencing reactions were performed and sequences determined automatically in a genetic analyzer (1<sup>ST</sup> Base, Malaysia) using the PCR primers mentioned above. Sequences obtained in this study were compared to those from GenBank database using the BLAST software on the NCBI website: (<http://www.ncbi.nlm.nih.gov/BLAST/>). After multiple alignment of selected sequencer with Clustal X. Phylogenetic trees were constructed using the PUAP beta 10 software version 4.0 (Swofford 2002).

## Results

### Taxonomic description

***Muscodor cinnamomi*** Suwannarach, K.D. Hyde & Lumyong, **sp. nov.**      FIGS 1–5

MYCOBANK # MB518008, GENBANK # GQ848369

*Fungus in natura cum Cinnamomi bejolghota consociatus et est deuteromycete myceliis sterilibus pertinens. Coloniae fungales est luteus in vitro examinati in loco cum sol lux. Sporae vel corpora fructificantia substatibus ullis non observata. Hyphae (0.9–5.2 µm) vulgo ramificantes et convolventes, fila stripformia et spiras perfectas (4.5–12 µm) formantes. In vitro examiniter corpores colifloriform (6.3–14 µm) e repletus forma hyphae.*

ETYMOLOGY: *cinnamomi*, from the name of the host plant.

HOLOTYPE: Thailand, Doi Suthep-Pui National Park; from a leaf of *Cinnamomum bejolghota* (Lauraceae), May 2008, Nakarin Suwannarach; **holotype** – dried culture, SDBR CMU-Cib 461. (Living culture, BCC38842).

TELEOMORPH: Unknown.

In nature, the fungus is associated with *Cinnamomum bejolghota* and it is an ascomycete with sterile mycelium. Fungal colonies whitish on all media (PDA, MA and CMA) when grown in darkness (FIG. 1), pale orange when grown in natural light. Hyphae (0.9–5.2 µm thick) commonly appearing as fused rope-like strands, branching (FIG. 4); with coils (4.5–12 µm diam.; FIGS 2, 3) and cauliflower-like bodies (6.3–14 µm; FIG. 5). Mycelium on PDA reaching 9 cm in 2–3 weeks and producing a fruity odor. Spores and other fruiting bodies did not develop under any conditions tested.

### Molecular phylogeny of *Muscodor cinnamomi* CMU-Cib 461

Partial ITS1 5.8 ITS2 rDNA sequences of *M. cinnamomi* were obtained and compared with GenBank database. After searching the ITS-5.8S rDNA sequences, 635 bp of *M. cinnamomi* (GQ848369) was subjected to an advanced BLAST search. The ITS1 5.8S ITS2 rDNA sequences of *M. cinnamomi* blasted five type strains of *Muscodor* species. The result showed that there was a 99, 99, 99, 98 and 90% similarity with *M. albus* (AF324336), *M. roseus* (AY034665), *M. crispans* (EU195297), *M. vitigenus* (AY100022) and *M. yucatanensis* (FJ917287), respectively.

Parsimony analysis of the alignment yielded 100 most parsimonious trees with total length of 873 steps (CI = 0.705, RI = 0.746, RC = 0.526, HI = 0.294), one of which is shown in FIG. 6. *Muscodor cinnamomi* and *Muscodor* species from GenBank formed a monophyletic clade (clade I) with a high bootstrap support (99%), and formed a sister group to *Anthostomella* (clade II) with 83% bootstrap support. *Muscodor* species are more closely related to the *Xylariaceae* than *Amphisphaeriaceae* with 100% bootstrap support.

### Volatile compounds from *M. cinnamomi* (CMU-Cib 461)

*Muscodor cinnamomi* (CMU-Cib 461) produced at least 11 volatile compounds. These could be positively identified on the basis of a GC/MS

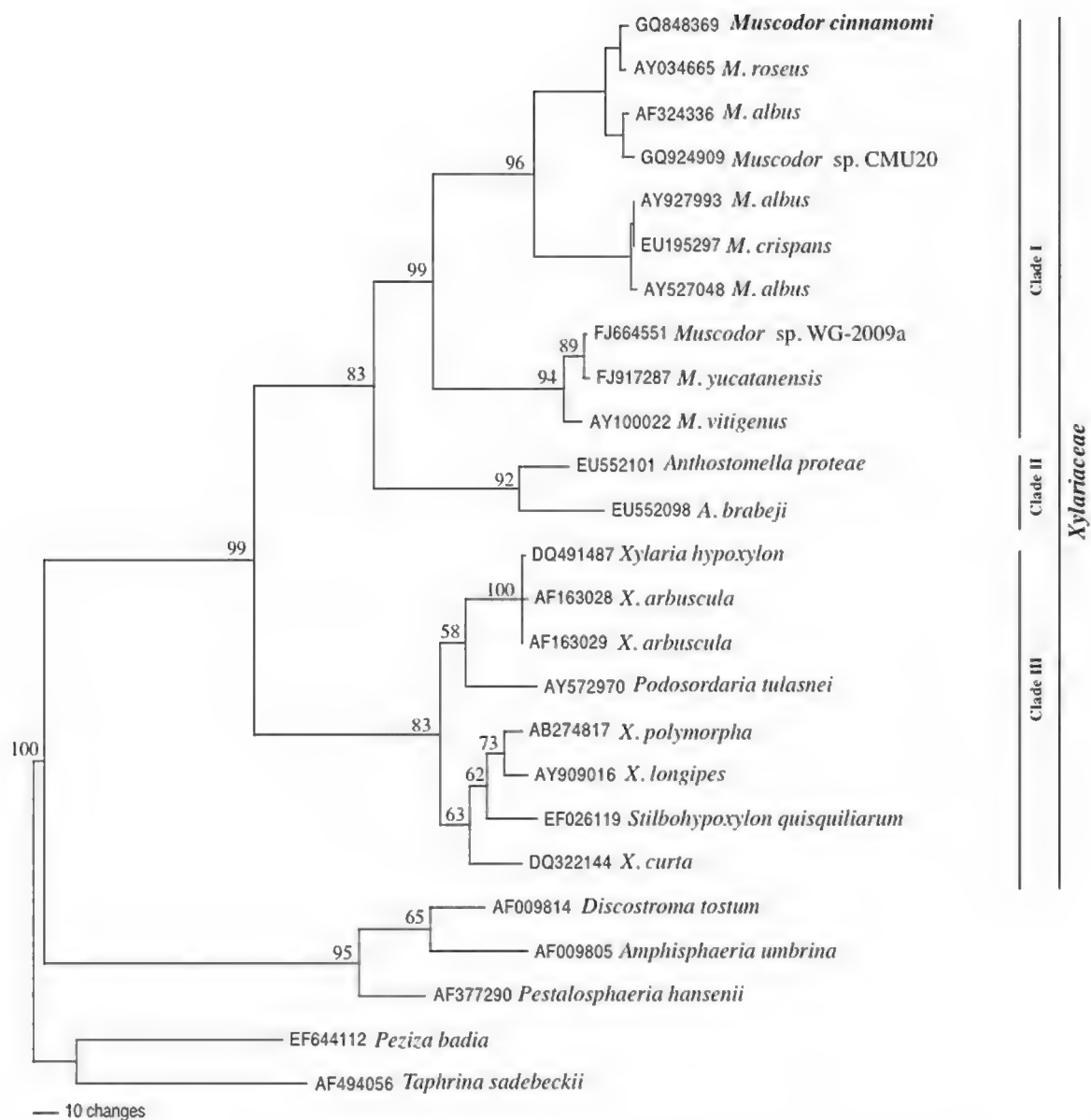


FIG 6. One of 100 most parsimonious trees inferred from a heuristic search of the ITS1-5.8S-ITS2 rDNA sequence alignment of 25 isolates of *Muscodor* and related genera. *Peziza badia* and *Taphrina sadebeckii* were used to root the tree. The size of the branches is indicated with a scale bar. Branches with bootstrap values  $\geq 50\%$  are shown at each branch.

comparison with authentic standards obtained from commercial sources as well as organic synthesis. The compounds were identified primarily on the basis of their mass spectral properties when compared to the NIST database. Of the compounds produced by this organism the most abundant were propanoic acid, 2-methyl, methyl ester, butanoic acid, 2-methyl, methyl ester and cis-2,4-dimethylthiane,S,S-dioxide with total area higher than 10% (TABLE 1). A number of other volatiles appeared that were unique to this isolate, including cis-2,4-dimethylthiane,S,S-dioxide;  $\beta$ -humolene; cyclopentane; eudesma4(14),11-diene and 1,1,1,5,7,7,7-heptamethyl-3,3-bis(trimethylsiloxy) tetrasiloxane compounds. In addition, the fungus produced azulene, but no naphthalene compounds.

TABLE 1. GC/MS analysis of the volatile compounds produced by *Muscodor cinnamomi* (CMU-Cib 461) culture in 5.0 mL clear glass vial Agilent® for 10 days.

RT (min:s)	TOTAL AREA (%)	ANALYSIS COMPOUND	M/z
3:15	1.10	(S)-(+)-5-methyl-1-heptanol	130
3:32	5.49	ethyl acetate	88
4:35	32.26	propanoic acid,2-methyl,methyl ester	102
5:38	11.35	cis-2,4-dimethylthiane,S,S-dioxide*	162
5:41	7.69	cyclopentane*	70
6:38	14.90	butanoic acid,2-methyl,methyl ester	116
9:29	3.12	1-butanol,3-methyl,acetate	130
27:42	3.23	β-humulene*	204
30:89	8.58	azulene, 1,2,3,5,6,7,8,8a-octahydro-1,4-dimethyl-7-(1-methylethenyl)-,[1S-(1.α., 7.α., 8a.β)]	204
30:90	7.32	Eudosma-4(14),11-diene*	107
34:48	2.66	1,1,1,5,7,7,7-heptamethyl-3,3-bis(trimethylsiloxy) tetrasiloxane	444

Abbreviations: \* = compounds found in *M. cinnamomi* but not in other *Muscodor* species;  
RT = retention time; M/z = mass to charge ratio.

## Discussion

*Muscodor cinnamomi* is introduced as a new species based on differences in colony characteristics, growth rate, ITS sequence data and volatile compounds produced. *Muscodor cinnamomi* (CMU-Cib 461) produced a white mycelium on a PDA. Spores or fruiting structures did not develop on any media including ones containing the host plant material, cinnamon leaves. In this respect it is similar to other *Muscodor* species. The hyphae tend to intertwine to form rope-like strands. Other species of *Muscodor* also have this tendency (Worapong et al. 2001, 2002). The fungus also produces cauliflower-like structures, which is similar to *M. crispans*. The features of *M. cinnamomi* (CMU-Cib 461) are similar to *M. albus*, *M. crispans*, *M. vitigenus* and *M. yucatanensis* which produce whitish mycelium on all media tested in artificial light (Worapong et al. 2001, Daisy et al. 2002, Mitchell et al. 2008, González et al. 2009). *Muscodor cinnamomi* developed a pale orange coloured mycelium in natural light, while *M. crispans* produces a pale pink mycelium in natural light (Mitchell et al. 2008). Phylogenetic analysis of the sequences of ITS1, 5.8S, and ITS2 showed that *M. cinnamomi* was closely related the other *Muscodor* species, which are related to family *Xylariaceae* (Worapong et al. 2001, 2002).

When measured by GC/MS, the fungus consistently produced alcohols, esters and small molecular weight acids, in the gas phase, when grown on PDA. *Muscodor cinnamomi* produces propanoic acid,2-methyl,methyl ester, which is similar to other *Muscodor* species. However, there are differences in other



TABLE 2. Synopsis of azulene and naphthalene production\* by *Muscodor* species.

SPECIES	AZULENE	NAPHTHALENE	DATA SOURCE
<i>M. albus</i>	+	+	Worapong et al. 2001
<i>M. cinnamomi</i>	+	–	This paper
<i>M. crispans</i>	–	–	Mitchell et al. 2008
<i>M. roseus</i>	+	+	Worapong et al. 2002
<i>M. vitigenus</i>	+	+	Daisy et al. 2002
<i>M. yucatanensis</i>	n	–	González et al. 2009

\* (+) = production; (–) = non-production; (n) = unreported.

compounds produced by the different *Muscodor* species (TABLE 2). The volatile compounds showed inhibition ability and lethal activity against a number of plant and human pathogens (Strobel et al. 2001, Worapong & Strobel 2009). Details on the bioactivities of this interesting genus appear elsewhere (Worapong et al. 2001, 2002, Daisy et al. 2002, Ezra et al. 2004, Strobel 2006, Strobel et al. 2007, Mitchell et al. 2008). The strain CMU-Cib 461 shared all of the common features of previously described *Muscodor* species but there were a number of different aspects to the taxon that distinguished it from other *Muscodor* species.

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## MYCOTAXON

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***Paecilomyces echinosporus* sp. nov.,  
a species isolated from soil in China**MINGJUN CHEN<sup>1</sup>, NA ZHOU<sup>1</sup>, ZENGZHI LI<sup>1</sup>, GI-HO SUNG<sup>2\*</sup> & BO HUANG<sup>1\*</sup>*chenmingjun2007@yahoo.cn zhouna0116@yahoo.com.cn**zzli@ahau.edu.cn bhuang@ahau.edu.cn*<sup>1</sup> Anhui Provincial Key Laboratory of Microbial Control  
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**Abstract** — During a survey of entomopathogenic fungi in China, a new species of *Paecilomyces* was isolated from a soil sample collected from Anhui province in China. It is differentiated from previously described species based on the morphology of its minutely echinulate conidia and conidiophores that possess penicillate phialides. Phylogenetic analyses with ITS region indicate that it is distantly related to *Isaria* and a close relative of *P. carneus*. The new species, *Paecilomyces echinosporus*, is presented with its Latin diagnosis, English description, and illustration. The type isolate and holotype are deposited in the Research Center for Entomogenous Fungi of Anhui Agricultural University (RCEF).

**Key words** — taxonomy, morphological characteristics, molecular identification

**Introduction**

The genus *Paecilomyces* was established by Bainier in 1907 and differentiated from the genus *Penicillium* Link by its colony that lacks green color, cylindrical conidiogenous cells, and the slime mass of spores (Samson 1974). The generic concept of *Paecilomyces* was later expanded to include species of genera *Isaria* and *Spicaria* that possess a conidiogenous structure similar to that of *P. variotii*, the type species of *Paecilomyces* (Brown & Smith 1957). The most comprehensive monographic work (Samson 1974) divides *Paecilomyces* species into two sections (i.e., *P. sect. Paecilomyces* and *P. sect. Isarioidea*) based on their teleomorphic affinities, colony color, odor, and growth temperature.

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Conducting phylogenetic analyses based on the small rDNA subunit to arrive at a natural classification of *Paecilomyces*, Luangsa-ard et al. (2004) showed that *Paecilomyces* is polyphyletic and represents two distantly related classes (i.e., *Sordariomycetes* and *Eurotiomycetes*). As a result, *P. sect. Isarioidea* was revised taxonomically with the lectotypification and formal conservation of the generic name, *Isaria* (Gams et al. 2005, Hodge et al. 2005). Following phylogenetic analyses of *P. sect. Isarioidea* using the  $\beta$ -tubulin gene and ITS region, ten species of *P. sect. Isarioidea* were transferred to *Isaria*.

Liang et al. (2005) reviewed 32 known species of *Paecilomyces* in China, where 12 novel *Paecilomyces* species were reported based on the survey of soil-borne filamentous fungi from 2003–06 (Liang et al. 2009). Of these, six monophialidic species were transferred to a new genus, *Taifanglania*, based on their morphological characteristics and molecular analyses. In this study, we report a new species of *Paecilomyces* that was found during a survey of entomopathogenic fungi in soil in Anhui province, China. The morphological examination and phylogenetic analysis revealed a species with features that differed from previously described *Paecilomyces* species and was distantly related to some *Isaria* taxa. This new species is described below as *Paecilomyces echinosporus*.

## Materials and methods

### Sample collection and strain isolation

Strain RCEF4111 was isolated from soil samples collected from Qimen, Anhui province, China. A 5 g sample of soil was mixed with 100 ml of sterile distilled water containing 0.05% (v/v) Tween 80. The soil suspension was diluted to a concentration of  $10^{-2}$  after shaking for approximately four hours. A 200  $\mu$ l of the soil suspension was plated on one plate with the D0C2 selective medium (Shimazu & Sato 1996), and incubated at 25°C for approximately 5 days until the colonies were formed. Colonies that formed conidiogenous structures were transferred to SDAY (Sabouraud's dextrose agar with yeast) slants.

### Strain identification

Strain RCEF4111 was transplanted onto Czapek agar, potato dextrose agar (PDA), and Sabouraud's agar according to Brown & Smith (1957) and Samson (1974), and then was incubated at 25°C for 14 days. The isolated fungus was examined using classical mycological techniques based on growth rate, as well as macroscopic and microscopic characteristics. The strain was also tested to investigate its ability to grow on PDA at 35°C. The type strain, RCEF4111 (dried RCEF4111-DPC1, holotype), has been deposited in the Research Center for Entomogenous Fungi (RCEF), Anhui Agriculture University, China.

### DNA extraction

For DNA extraction, spores were inoculated to Petri dish containing SDAY medium overlaid with a disc of sterilized cellophane. After incubating at 25°C for approximately

TABLE 1. Accession numbers, strain numbers, and origins of *Paecilomyces* spp. and other taxa used for phylogenetic analysis.

GENBANK #	NAME	STRAIN #	REFERENCES
AJ786573	<i>Cordyceps militaris</i> (L.) Link	3856.H.	Stensrud et al. (2005)
AY624168	<i>Isaria amoenerosea</i> Henn.	CBS 107.73 T	Luangsa-ard et al. (2005)
AY624172	<i>I. cateniannulata</i> (Z.Q. Liang) Samson & Hywel-Jones	CBS 152.83	Luangsa-ard et al. (2005)
AY624175	<i>I. cicadae</i> Miq.	BCC 2574	Luangsa-ard et al. (2005)
AY624176	<i>I. coleopterorum</i> (Samson & H.C. Evans) Samson & Hywel-Jones	CBS 102.73	Luangsa-ard et al. (2005)
AY624181	<i>I. farinosa</i> (Holmsk.) Fr.	CBS 111113	Luangsa-ard et al. (2005)
AY624184	<i>I. fumosorosea</i> Wize	CBS 107.10	Luangsa-ard et al. (2005)
AY624186	<i>I. javanica</i> (Frieder. & Bally) Samson & Hywel-Jones	CBS 134.22	Luangsa-ard et al. (2005)
AY624196	<i>I. tenuipes</i> Peck	ARSEF 5135	Luangsa-ard et al. (2005)
AY624202	<i>Mariannaea camptospora</i> Samson	CBS 209.73	Luangsa-ard et al. (2005)
AF135210	<i>Metarhizium anisopliae</i> (Metschn.) Sorokin var. <i>anisopliae</i>	FI1029	Driver et al. (2000)
AF368270	<i>M. cylindrosporum</i> Q.T. Chen & H.L. Guo	ACCC 30114 T	Huang et al. (2004)
AF138270	<i>M. flavoviride</i> W. Gams & Rozsypal var. <i>flavoviride</i>	FI 38	Driver et al. (2000)
AF368501	<i>Nomuraea rileyi</i> (Farl.) Samson	RCEF 0292	Huang et al. (2004)
AY624170	<i>Paecilomyces carneus</i> (Duché & R. Heim) A.H.S. Br. & G. Sm.	CBS 399.59	Luangsa-ard et al. (2005)
AY624174	<i>P. cinnamomeus</i> (Petch) Samson & W. Gams	CBS 398.86	Luangsa-ard et al. (2005)
GU108582	<i>P. echinosporus</i> Ming J. Chen, G.H. Sung & B. Huang	RCEF 4111	In this study
AJ536552	<i>P. gunnii</i> Z.Q. Liang	ZSU 20872	Unpublished
AY624189	<i>P. lilacinus</i> (Thom) Samson	CBS 284.36 T	Luangsa-ard et al. (2005)
AY624193	<i>P. marquandii</i> (Masse) S. Hughes	CBS 182.27 T	Luangsa-ard et al. (2005)
AY624192	<i>P. niphedodes</i> Samson	CBS 364.76	Luangsa-ard et al. (2005)
AY624194	<i>P. penicillatus</i> (Höhn.) Samson	CBS 448.69	Luangsa-ard et al. (2005)
AY624197	<i>P. viridis</i> Segretain et al. ex Samson	CBS 348.65	Luangsa-ard et al. (2005)
EU004811	<i>Taifanglania curticatenata</i> (Z.Q. Liang & Y.F. Han) Z.Q. Liang et al.	HC 125-2 T	Liang et al. (2009)

7 days, genomic DNA was extracted from the mycelia scraped from the cellophane using benzyl chloride (Zhu et al. 1994). The extracted DNA was stored in 100 µL TE buffer (10mM Tris-HCl, PH8.0; 1mM EDTA) at 4°C, and was diluted 10-fold with TE buffer for the following PCR reactions.

PCR amplification and determination of ITS sequencing

The PCR amplification of ITS region was performed using the primers of ITS5 and ITS4 (White et al. 1990). The PCR conditions are as follows: 94°C for 5 mins, 35 cycles of 94°C for 1 min, 52°C for 1 min, 72°C for 2 mins and 72°C for 10 mins. The PCR reaction



was conducted in 25  $\mu\text{L}$  volume with the following components: 2.5  $\mu\text{L}$  of 10  $\times$  reaction buffer, 0.5  $\mu\text{L}$  of each dNTP, 1  $\mu\text{L}$  of each primer, and 2 units of Taq DNA polymerase, 2  $\mu\text{L}$  of the diluted DNA and 16.8  $\mu\text{L}$  of ddH<sub>2</sub>O. The resulting PCR product was examined on 1.2% TBE agarose gel stained with ethidium bromide. After purifying PCR product using EasyPure quick gel extraction kit (TransGen Biotech), DNA sequencing was carried out at Sangon Company (Shanghai, China) and the resulting ITS sequence of RCEF 4111 was submitted to GenBank with accession number GU108582.

### Sequence alignment and phylogenetic analysis

DNA sequences that are generated in this study and downloaded from GenBank were aligned using Clustal X 1.81 (Thompson et al. 1997). The alignment was manually adjusted to maximize homology. Maximum parsimony analyses were conducted using PAUP\* 4.0b10 (Swofford 2002) with 1,000 replicates of heuristic search of random sequence additions, branch swapping by tree bisection-reconnection (TBR) and MulTrees in effect. In the parsimony analyses, unambiguously aligned gaps were treated as a new state and all characters were equally weighted. Branch support was estimated by bootstrapping using 1,000 replicates of 10 replicates of heuristic search with the same option (Felsenstein 1985). We also performed a BLAST search with the obtained sequence of the new taxon as a query to find the close relatives in GenBank database.

## Results

### Taxonomy

*Paecilomyces echinosporus* Ming J. Chen, G.H. Sung & B. Huang, sp. nov. FIG. 1  
MYCOBANK 518113; GENBANK GU108582

*Coloniae in agaro Czapekii ad 30–37 mm diam post 14 dies 25°C, in medio modice sulcatae, albae, pulverulentae, margine regulari; reversum luteolum; 35°C haud crescit. Hyphae vegetativae hyalinae, septatae, ramosae, leves, 2.0–3.5  $\mu\text{m}$  latae. Apparatus conidialis elongatus vel compactus, seu phialides singulae seu capitula verticillos ramorum et phialidum ferentia; stipites ex hyphis aeriis orientes, vulgo 45–95  $\times$  2.5  $\mu\text{m}$ . Phialides ad quinae verticillatae, 9.5–15.5  $\times$  2.0–3.0  $\mu\text{m}$ , e basi cylindrica et collulo angusto minus quam 0.5  $\mu\text{m}$  lato composita. Conidia unicellularia, minute echinulata, subglobosa vel ellipsoidea, 2.7–5.0  $\times$  2.0–3.0  $\mu\text{m}$ . Chlamydosporae absentes.*

**HOLOTYPE** — RCEF4111 was isolated by B. Huang & N. Zhou from soil of Qimen, Anhui province, China, in March, 2008, deposited in the Research Center for Entomogenous Fungi (RCEF).

Colony on Czapek agar attaining a diameter of 30 to 37 mm within 14 days at 25°C, slightly ridged at the center, white, powdery, regular in the margin; reverse yellowish. Colony growth not observed at 35°C. Vegetative hyphae hyaline, septate, branched, smooth-walled, 2.0–3.5  $\mu\text{m}$  wide. Conidial structures elongated to compact, varying in complexity from single detached phialides to heads with a terminal whorl of phialides and whorl of branches, conidiophores arising from aerial hyphae, normally 45–95  $\times$  2.5  $\mu\text{m}$ . Phialides up to 5 in a whorl, 9.5–15.5  $\times$  2.0–3.0  $\mu\text{m}$ , consisting of a cylindrical basal portion, tapering into a thin neck, less than 0.5  $\mu\text{m}$  wide. Conidia one-celled, minutely echinulate, subglobose to ellipsoidal, 2.7–5.0  $\times$  2.0–3.0  $\mu\text{m}$ . Chlamydospores absent.

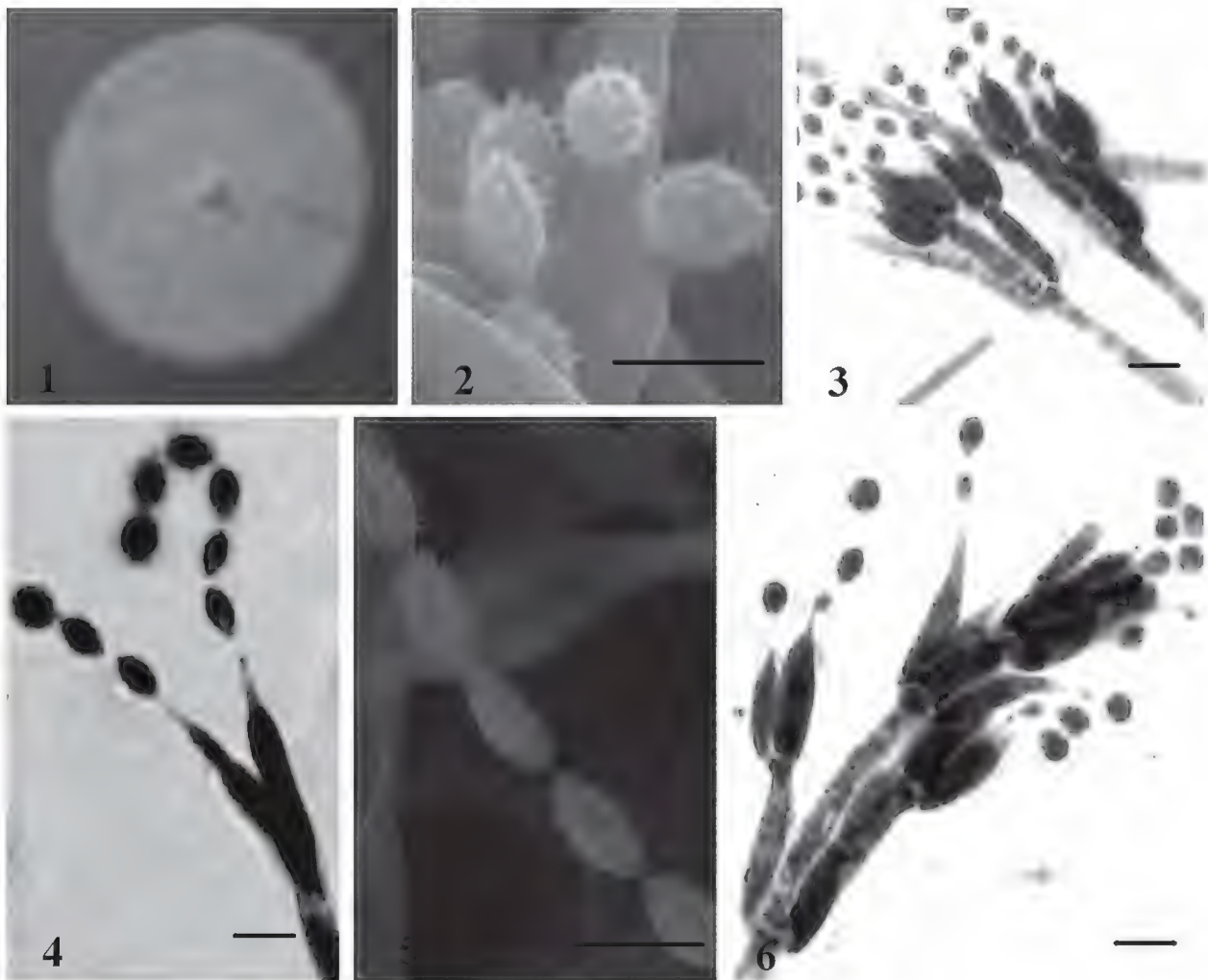


FIG. 1. Colony and conidiogenous structure of *Paecilomyces echinosporus* (Bars = 5 $\mu$ m). 1—Colony on Czapek agar; 2, 5—echinulate conidia; 3,4, 6—phialides and echinulate conidia.

#### Molecular Characteristics of *Paecilomyces echinosporus*

The ITS (ITS1, 5.8S rDNA, and ITS2) region is 538 bp long. ITS dataset with 23 strains contains 732 characters including 259 parsimony-informative characters. The single tree generated from maximum parsimony (TL= 1078, CI= 0.5965, HI = 0.4035, RI = 0.6432, RC = 0.3836) is shown in FIG. 2. The phylogenetic tree inferred from the ITS sequence data clusters isolate RCEF4111 with *P. carneus* with 95% bootstrap support. In addition to the phylogenetic analysis, we performed a BLAST search with ITS sequence of *P. echinosporus* as a query. Search results imply that *P. echinosporus* is most comparable to *P. marquandii* (ARSEF 3047, EU553322, 97%), *P. lilacinus* (CG 348, EU553317, 97%), and *P. carneus* (CBS 399.59, AY624170, 90%). A NCBI BLAST search yielded a sequence max identity of the *P. echinosporus* ITS sequence of 100% with Malian strain ARSEF 3047 and Brazilian strain CG 348 and showed the closest relative of these two isolates as *P. carneus* (GC 525, EU553292, 91%). Therefore, ARSEF 3047 and CG 348 appear either closely related to or conspecific with *P. echinosporus*, indicating the presence of the species in Brazil and Mali.

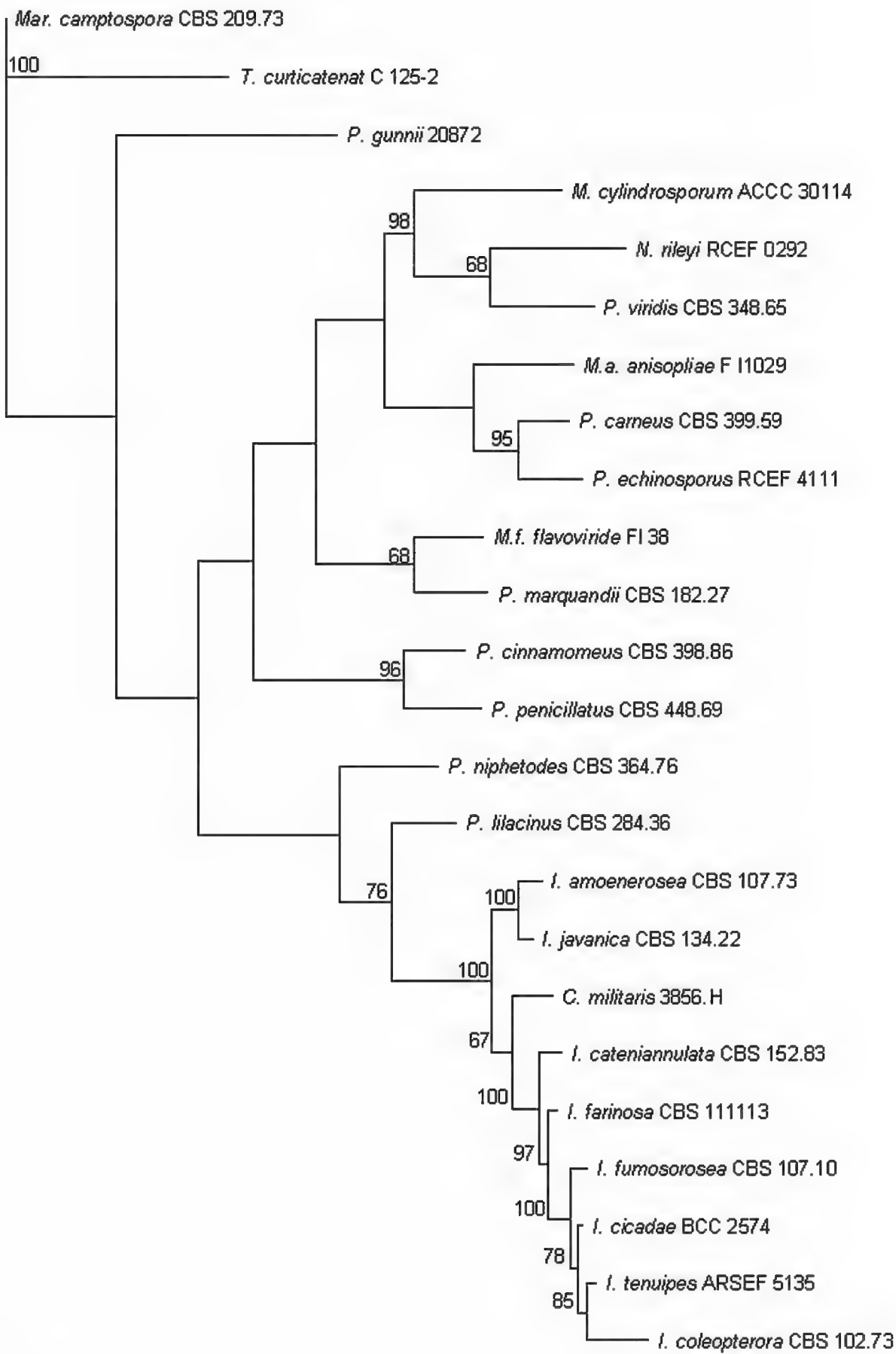


Fig. 2. Phylogenetic tree generated from parsimony analysis based on ITS rDNA sequences. Numbers at the nodes give bootstrap support derived from 1000 replicates. *Mariannaea camptospora* was used as outgroup.

## Discussion

In *Paecilomyces*, based on the morphological characters, species that produce echinulate or rough conidia include *P. carneus*, *P. gunnii*, *P. marquandii*, and *P. lilacinus* (Samson 1974, Liang 1985, Han et al. 2005). Although the conidia are echinulate in both *P. carneus* and *P. gunnii*, they can be differentiated by the color of the reverse side of the colony in culture; *P. carneus* is dark green, while *P. gunnii* produces a dark brown colony and chlamydospores. Meanwhile, conidia are rough in *P. marquandii* and *P. lilacinus* but possess purple or vinaceous conidial heads. In addition, Chlamydospore-like cells are usually present in *P. marquandii* and *P. lilacinus* conidiophores are pigmented and rough-walled, while *P. echinosporus* does not produce chlamydospores and possesses white and smooth conidiophores.

Our phylogenetic analysis of *Paecilomyces* species clusters *P. echinosporus* and *P. carneus* together in a clade and distinctly related to the other four species that produce echinulate or rough conidia (FIG. 2). Although the new species resembles *P. carneus* in the echinulate conidia, *P. echinosporus* and *P. carneus* share only 91% sequence similarity. In morphological comparison, *P. echinosporus* produces conidiophores with penicillate branches and short-necked phialides and a white colony with a yellow reverse. In contrast, *P. carneus* produces conidiophores with verticillate branches and phialides that taper into a thin long neck and a pink (after sporulation) colony with a mostly green to dark green reverse. Our combined traditional morphological study and molecular analyses identify strain RCEF4111 isolated from soil sample as a new species of *Paecilomyces*, *P. echinosporus*.

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***Lolia aquatica* gen. et sp. nov.  
(*Lindgomycetaceae*, *Pleosporales*), a new coelomycete from  
freshwater habitats in Egypt**

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**Abstract** — An unknown coelomycete that was collected from the River Nile and associated irrigation canals in Egypt is described. The fungus is characterized by gelatinous pearl white acervuli, a peridium that forms textura intricata, holoblastic conidia that have one basal excentric cellular appendage, and up to 3–5 sub-apical cellular attenuating appendages. Based on morphology, no described genus can accommodate this new fungus, so it is described herein as new genus and species. Phylogenetic analyses of the 28S ribosomal large subunit (LSU) rDNA sequence placed the new fungus in the family *Lindgomycetaceae*, *Pleosporales*, *Dothideomycetes*.

**Key words** — aquatic fungi, anamorphic fungi, subtropical, appendaged conidia

## Introduction

Over 7000 coelomycetes in 1000 genera (+ 500 syn.) have been described (Kirk et al. 2008) from a wide range of substrates and geographical locations (Sutton 1980, Nag Raj 1993). A small number of coelomycetes have been linked to their teleomorphs, with affinities to ascomycetes, while a few are basidiomycetes (Nag Raj 1978, 1980, Dyko & Sutton 1979, Cole & Samson 1979, Nag Raj et al. 1989, Rungjindamai et al. 2008). Coelomycetes are a major group of the aquatic mycota of *Phragmites australis* (Van Ryckegem & Verbeken 2005a,b, 2007; Abdel-Aziz 2008). During an investigation of aquatic fungi in Egypt an unknown coelomycete with gelatinous pearl white acervuli was recorded from different localities at the River Nile and irrigation canals in Upper Egypt. This fungus is unique in that it possesses one excentric basal and three to five sub-apical un-branched cellular appendages of type A (Nag Raj 1993). This newly

discovered taxon is described, illustrated, and compared to other appendaged coelomycetes. In addition, we used phylogenetic analyses of the LSU gene to determine its phylogenetic relationship.

## Materials and methods

### Collection of the fungi

Submerged decayed wood was collected from the River Nile and irrigation canals from Sohag, Qena, and Aswan governorates. Samples were kept in clean plastic bags and returned to the laboratory, examined immediately under stereomicroscope for fungal fruiting structures and subsequently incubated on moist filter paper in sterile plastic boxes. Material was examined periodically over three month's incubation. Single spore isolates of the new fungus were obtained. Photographs were taken using an Olympus BX51 differential interference contrast light microscope and Olympus DP12 digital imaging system (Olympus Corporation, Tokyo, Japan). Herbarium material was dried at 60°C for 24 h and deposited along with the isolated fungal cultures in the authors' culture collection, Department of Botany, Faculty of Science, Sohag University, Egypt. Voucher slides and type material of the new fungus were deposited at International Mycological Institute (IMI).

### DNA extraction, sequencing, and phylogenetic analysis

Single-spore isolate of the fungus was grown in YMG broth (4 g yeast extract, 10 g glucose, 10 g malt extract in 1 liter distilled water) until sufficient mycelium had formed to allow DNA extraction. DNA extraction for polymerase chain reaction (PCR) was performed using the Microbial DNA Extraction Kit (MOBIO; Mo Bio Laboratories, Carlsbad, CA, USA) according to the manufacturer's instructions. Partial LSU ribosomal DNA was amplified using primers LR0R and LR7 (Bunyard et al. 1994). PCR reactions, cycling parameters and sequencing were carried out as described by Abdel-Wahab et al. (2009). Sequences were assembled using Sequencher 4.2.2 (Gene Codes Corporation). Sequences were aligned with others retrieved from GenBank using ClustalX (Thompson et al. 1997) and optimized manually. The positions where one or more species contained a length mutation and ambiguously aligned regions were not included in the subsequent phylogenetic analysis. Nucleotide sequence phylogenies were constructed using PAUP\* 4.0b10 (Swofford 2002). Maximum-likelihood (ML) analyses (Felsenstein 1981) were performed using heuristic searches with the random stepwise addition of 100 replicates and tree bisection-reconnection (TBR) rearrangements. The optimal model of nucleotide substitution for the ML analyses was determined using hierarchical likelihood ratio tests as implemented in Modeltest 3.7 (Posada and Crandall 1998). The model selected as the best fit for LSU rDNA data set was TrN+I+G. For the bootstrap analyses (Felsenstein 1985), 100 replicates were generated with 5 random additions and TBR. Maximum-parsimony (MP) trees were obtained by 100 random addition heuristic search replicates using PAUP, and 1000 bootstrap replicates were performed employing 5 random addition heuristic searches. Posteriori probability values were obtained by using the MrBayes 3.1.2 (Huelsenbeck & Ronquist 2001, Ronquist & Huelsenbeck 2003) with the SYM+I+G model that was determined using MrModeltest 2.2 (Nylander 2004). Five million generations were run in four chains with sampling every 100 generations,

yielding 50 000 trees, of which the first 12 500 were discarded as “burn in.” The numbers on the branches are estimates of a posteriori probabilities. The LSU sequence of *Lolia aquatica* isolate used in this study was deposited at GenBank under the accession number “HM367732”; ex-type: MF 644 (JAMSTEC, Japan).

Results

Phylogenetic analyses

The partial LSU rDNA sequence of *Lolia aquatica* is aligned with representatives of the family *Lindgomycetaceae* along with representatives of the families belong to orders *Pleosporales* and *Jahnulales*. In total, the LSU rDNA dataset include 39 taxa of which 2 belong to the class *Pezizomycetes* that

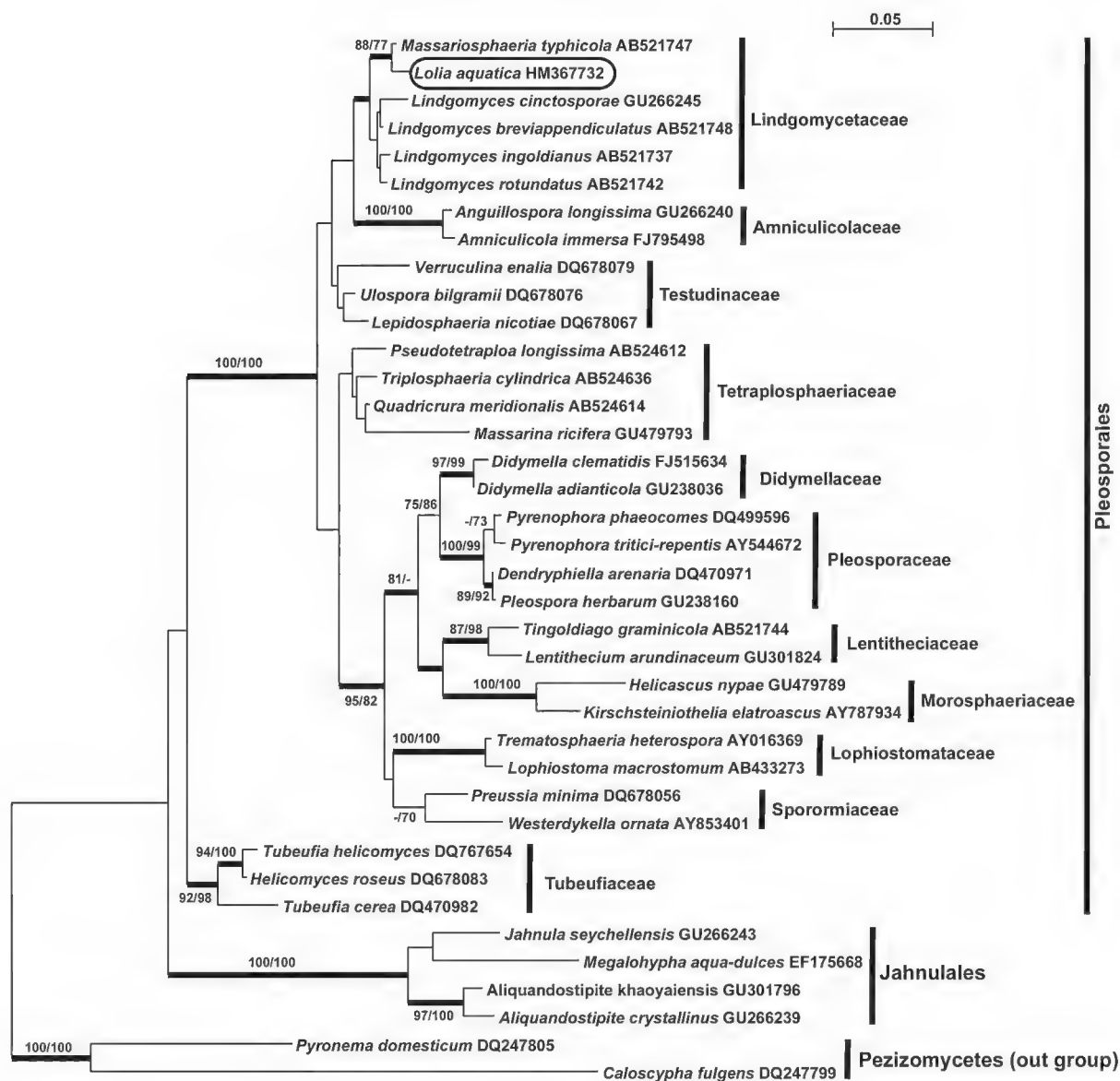


FIG. 1: Phylogenetic relationships of *Lolia aquatica* and closely similar fungi, based on the nucleotide sequences of the large subunit (LSU) rDNA. The maximum likelihood tree (ML) (–ln likelihood = 4978.2339) was constructed as described in the text. The numbers indicate pp values ≥ 95% (in bold), ML bootstrap and MP bootstrap values ≥ 70%. The new species, *Lolia aquatica*, is highlighted in a box.

was used as outgroup. The dataset consisted of 795 total characters, of which 42 gaps are excluded, 477 characters were constant, 69 variable characters were parsimony-uninformative and 207 were parsimony informative characters. Five most parsimonious trees were produced using heuristic search, the five trees have equal length of 817 steps, a consistency index of 0.5141, a retention index of 0.6953 and a rescaled consistency index of 0.3574. Maximum likelihood analysis produced one tree with  $-\ln$  likelihood score of 4,978.2339 (FIG. 1). Most parsimonious (MP), and Neighbor-Joining (NJ) and Bayesian analyses produced similar trees to the one shown in FIG. 1.

*Lolia aquatica* is a sister taxon to *Massariosphaeria typhicola* (P. Karst.) Leuchtm. and forms a well supported clade (100/88/77 for Bayesian/ML/MP respectively) within the recently published freshwater ascomycete family, *Lindgomycetaceae* K. Hiray et al. (Schoch et al. 2009, Shearer et al. 2009, Hirayama et al. 2010).

## Taxonomy

### *Lolia* Abdel-Aziz & Abdel-Wahab, **anam. gen. nov.**

MYCOBANK MB 518528

*Conidiomata* acervularia, margariticoloria, in gelatina immerse, superficialia, solitaria vel gregaria. Peridium ex textura intricata formatum, hyalinum, in matrice gelatinosa immersum. Conidiogenesis holoblastica. Conidia aseptata, clavata, cylindrica vel ellipsoidea, hyalina, levia, tenuitunicata, ad apicem 3–5 appendicibus, ad basim appendice singulari excentrica (typi A).

**TYPE SPECIES:** *Lolia aquatica* Abdel-Aziz & Abdel-Wahab

**ETYMOLOGY:** From the Arabic word, *Loli* = pearl, in reference to the color of the conidiomata.

*Conidiomata* acervular, superficial, pearl white, embedded in gel, single or aggregated. Peridium forming textura intricata, hyaline, embedded in gel. Conidiogenesis holoblastic. Conidia unicellular, clavate, cylindrical, ellipsoidal, hyaline, smooth, thin-walled, with basal and apical cellular, tapering, attenuating appendages of type A.

### *Lolia aquatica* Abdel-Aziz & Abdel-Wahab, **sp. nov.**

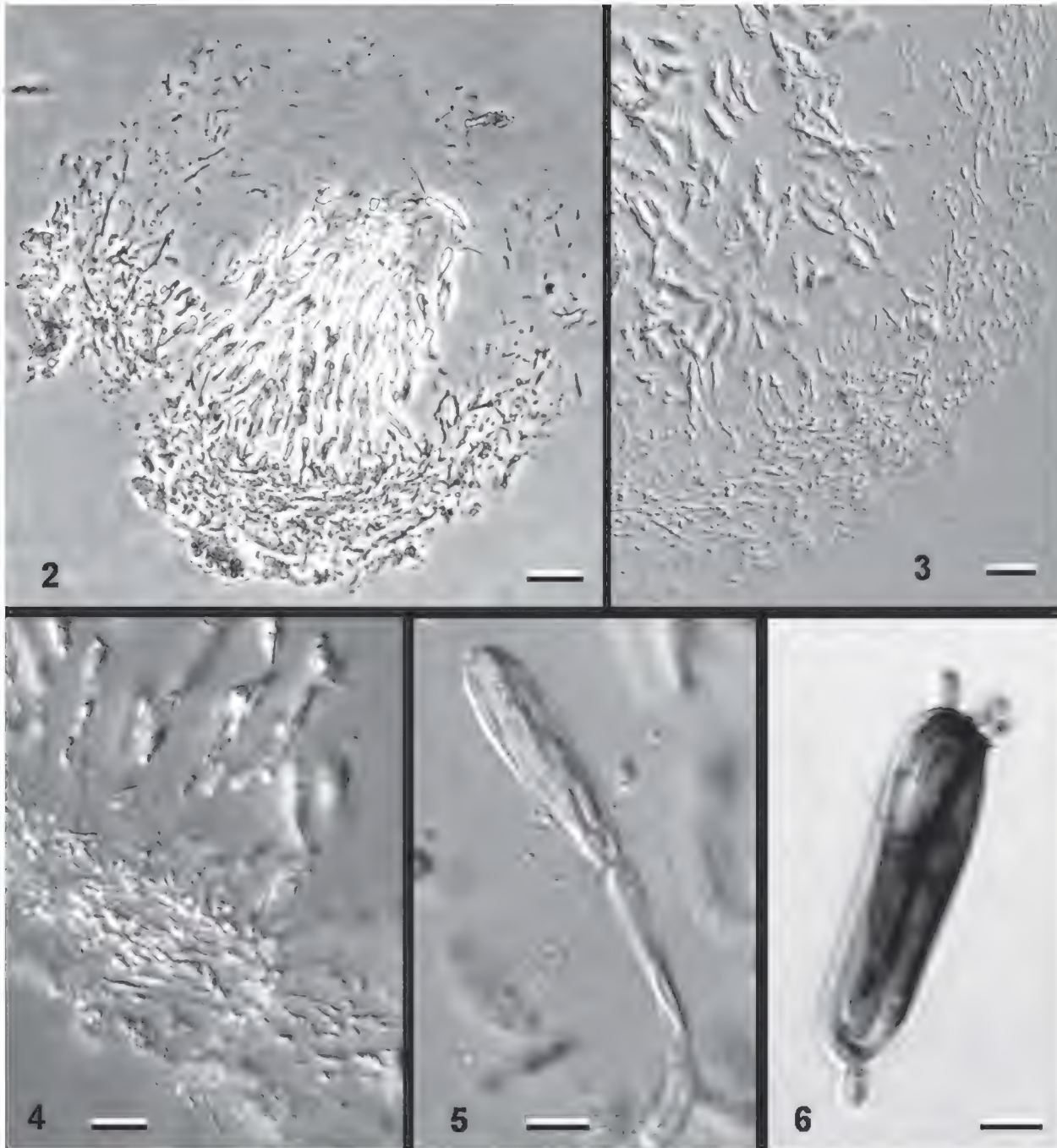
FIGS 2–10

MYCOBANK MB518529

*Conidiomata* acervularia, 400–480  $\mu\text{m}$  alta, 380–540  $\mu\text{m}$  diam., margariticoloria, superficialia, solitaria vel gregaria. Peridium 57–80  $\mu\text{m}$  crassum, ex textura intricata formatum, hyalinum, in matrice gelatinosa immersum. Conidiogenesis holoblastica. Conidia 31–45  $\times$  7–10  $\mu\text{m}$ , aseptata, hyalina, clavata, ellipsoidea vel cylindrical, 3–5 appendicibus apicalibus, 55–90  $\times$  1.5–3  $\mu\text{m}$ , et appendice basali singulari, simplici, excentrica, 10–85  $\times$  1.5–3  $\mu\text{m}$ .

**TYPE:** Egypt, Sohag, El Balyana city, on decayed stem of *Phragmites australis* (Cav.) Steud. at irrigation canal, March 2005, F.A. Abdel-Aziz (**Holotype**, IMI 398675; ex-type culture, MF644 (JAMSTEC, Japan); iso-type, MD644 (authors' culture collection).



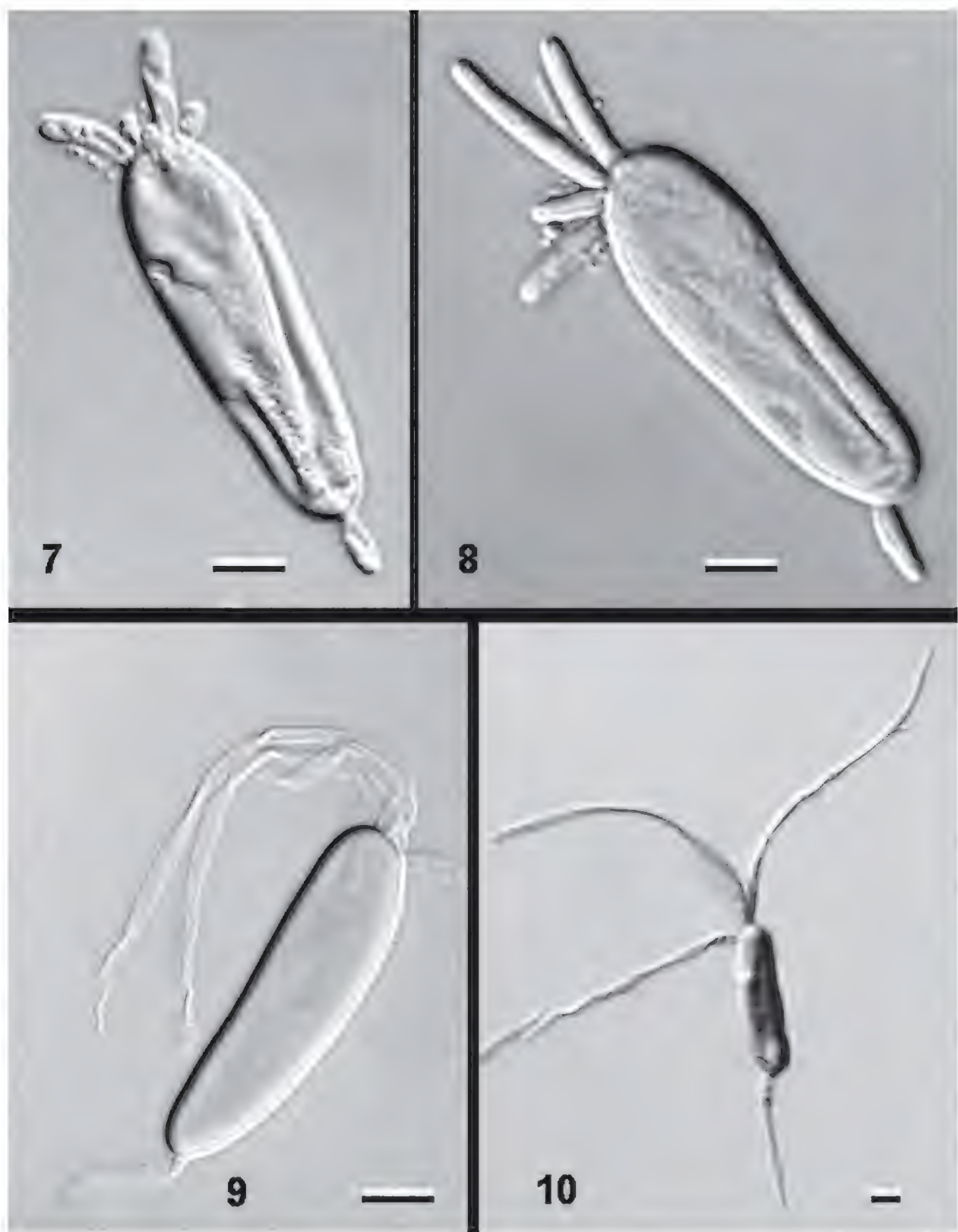


FIGS 2–6: *Lolia aquatica*. Differential interference contrast light micrographs (from holotype, mounted in water). 2. Vertical section through the gelatinous acervular (in phase contrast). 3–4. Magnified part of the peridial wall that forms textura intricata. 5. Young developing conidium at the tip of the conidiogenous cell. 6. Young conidium stained in toluidine blue shows initials of apical and basal appendages. Bars: 2 = 40  $\mu\text{m}$ , 3–4 = 20  $\mu\text{m}$ , 5–6 = 5  $\mu\text{m}$ .

ETYMOLOGY: From the Latin adjective *aquaticus*, in reference to the freshwater habitat of the fungus.

Conidiomata acervular, 400–480  $\mu\text{m}$  high, 380–540  $\mu\text{m}$  diam, pearl white when wet, dull yellow brown when dry, superficial, single or aggregated (FIG. 2). Peridium 57–80  $\mu\text{m}$  thick, forming textura intricata, hyaline, embedded in gel (FIGS 3–4). Conidiophores lining the acervuli wall and arising from innermost elements of the wall, loosely aggregated, branched and septate, colorless, smooth, embedded in gel. Conidiogenous cells cylindrical to sub-cylindrical,





FIGS 7–10: *Lolita aquatica*. Differential interference contrast light micrographs of conidia at different stages of development. 10. Stained in toluidine blue. Bars: 7–10 = 5  $\mu\text{m}$ .

colorless, smooth, bearing a single terminal conidium. Conidiogenesis: ontogeny holoblastic with apical wall building; delimitation by a transverse septum; secession schizolytic (FIG. 5). Conidia 31–45  $\times$  7–10  $\mu\text{m}$  (mean = 36  $\times$  8.6  $\mu\text{m}$ , n = 50), unicellular, hyaline, clavate, ellipsoidal, cylindrical, hyaline, smooth, thin-walled, solitary. Mean conidium length/width ratio = 4.2:1. Apical

appendages  $55\text{--}90 \times 1.5\text{--}3 \mu\text{m}$  (mean =  $68.6 \times 2.6 \mu\text{m}$ ,  $n = 20$ ), three to five sub-apical cellular appendages, attenuating, tapering. Basal appendage  $10\text{--}85 \times 1.5\text{--}3 \mu\text{m}$  (mean =  $27.9 \times 2.3 \mu\text{m}$ ), excentric, cellular, attenuating, tapering. Both apical and basal appendages are on one side of the conidia and arising as tubular extension of the conidium body and not separated from it at maturity by septa (FIGS 6–10).

## Discussion

Several groups of anamorphic fungi are present in freshwater habitats (Shearer et al. 2004, 2007). The best-known and the most studied group is the “aquatic” or “Ingoldian” hyphomycetes, which are distinguished by their tetra- or branched, or sigmoid conidia that are released into and dispersed by water (Ingold 1975, Webster & Descals 1981, Bärlocher 1992). About 300 species of aquatic hyphomycetes have been described thus far (Bärlocher 1992, Shearer et al. 2007). The “aeroaquatic hyphomycetes,” whose conidia are modified in a variety of ways to trap air for flotation, comprise a second group of anamorphic fungi (Fisher 1979, Michaelides & Kendrick 1982, Webster & Descals 1981, Premdas & Kendrick 1991). Coelomycetes are encountered regularly on a wide variety of submerged plant substrata in both lentic and lotic habitats (Shearer et al. 2004).

Phylogenetic analyses of partial 28S rDNA of *Lolia aquatica* show that it is a member of *Lindgomycetaceae*, *Pleosporales*. Phylogenetically, there are four major exclusive freshwater clades in the *Dothideomycetes* (Schoch et al. 2009), namely, the order *Jahnulales* (Pang et al. 2002, Campbell et al. 2007) and three recently described families: *Lindgomycetaceae*, *Amniculicolaceae* and *Lentitheciaceae* (Schoch et al. 2009, Shearer et al. 2009, Zhang et al. 2009, Hirayama et al. 2010).

There are several coelomycetous genera with hyaline, unicellular, appendages conidia that are somewhat similar to *Lolia aquatica*, e.g., *Chaetospermum* Sacc., *Giulia* Tassi, and *Mycotribulus* Nag Raj & W.B. Kend. *Lolia aquatica* is strikingly similar to *Chaetospermum* species, both having pearl white conidiomata, heavily gelatinized walls that consist of textura intricata, and conidia bearing type A appendages. However *Chaetospermum* species differ in having stromatic, pycnoid conidiomata, and an equal number of conidial appendages (3 to 6) at each end (Sutton 1980, Nag Raj 1993). Phylogenetic analyses of SSU and LSU rDNA placed *Chaetospermum* in the *Basidiomycota* (*Sebacinaceae*; Rungjindamai et al. 2008), whereas *L. aquatica* is in the *Ascomycota*.

The genus *Giulia* has dark-brown to black, immersed pycnidia, conidia bearing apical extra-cellular type D appendages arising by differential gelatinization of the conidium sheath. *Mycotribulus* has immersed to erumpent, brown pycnidia, filamentous paraphyses, and conidia bearing type A appendages at both sides

(one apical centric single appendage and 2-4 lateral basal appendages slightly above the truncate base). Phylogenetic analyses of SSU and LSU rDNA placed *Giulia* and *Mycotribulus* in the *Basidiomycota* (*Corticaceae* and *Physalacriaceae*, respectively; Rungjindamai et al. 2008).

There are several coelomycetous genera with septate hyaline or colored conidia with cellular apical and basal appendages: e.g., *Bartalinia* Tassi, *Discostroma* Clem., *Discosia* Lib., *Monochaetia* (Sacc.) Allesch., *Pestalotia* De Not., *Pestalotiopsis* Steyaert, *Seimatosporium* Corda, *Seiridium* Nees, *Truncatella* Steyaert. Phylogenetic analyses of LSU rDNA placed all the above-mentioned genera in the family *Amphisphaeriaceae*, *Xylariales* (Jeewon et al. 2002).

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# MYCOTAXON

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## ***Chlamydopsis*: an emendment of the genus and its type species**

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**Abstract** — *Chlamydopsis proliferans*, the type of a monotypic genus, was isolated on decaying leaves of *Caesalpinia echinata* in the Reserva Biológica de Mogi-Guaçu, São Paulo State, Brazil. During our study, we observed differences between our new collection and the original description. We therefore emend the circumscriptions of the genus and the species, which is reported for the first time from South America.

**Key words** — litter fungi, hyphomycetes, brazil-wood

### **Introduction**

During investigations of conidial fungi that occur on leaf litter of *Caesalpinia echinata*, brazil-wood (Silva & Grandi 2008), an interesting dematiaceous hyphomycete was isolated. The collection was identified as *Chlamydopsis proliferans* but showed distinct features different from the original description (Holubová-Jechová & Castañeda 1986).

*Chlamydopsis* is a monotypic genus, described from decaying leaves of *Lauraceae* in the Province of Camagüey, Cuba; since it was proposed there have been no other records nor have new species been added to the genus (Kirk et al. 2008, [www.indexfungorum.org](http://www.indexfungorum.org), consulted 14 June 2010). The conidia of our collection are typical and divided into two parts composed of one unicellular basal cell and an apical part with a central globose brown cell. Many delicate pale brown cells surround the central cell as illustrated by Holubová-Jechová & Castañeda Ruiz (1986), but in disagreement with their interpretation. Moreover the conidia are muriform since they possess septa in more than one plane (Kirk et al. 2008).

Therefore, emendments to the genus and species are proposed and the description and illustrations of the Brazilian material presented.

## Materials and methods

The leaf litter of *Caesalpinia echinata* was collected from February 2005 to February 2006 in the “Reserva Biológica de Mogi-Guaçu”, (22°15'02.4”S 47°09'28.9”W), São Paulo State, Brazil. After the dead leaves were successively washed, they were incubated in moist chambers at room temperature (Harley & Waid 1955, Grandi & Gusmão 1998). The fungal specimens were transferred to slide mounts prepared with lactophenol-cotton blue, polyvinyl alcohol, and glycerin (adapted from Morton et al. 1993, Mueller et al. 2004). Identification was made with microscope Axiostar plus and pictures with Axioskop 40, AxioCam MR and AxioVision, both Carl Zeiss. Permanent slides were deposited in the “Herbário Científico do Estado Maria Eneyda P. Kauffmann Fidalgo (SP)”, Brazil. In addition, the type specimen PRM 842703 (isotype) was requested from the Herbarium PRM, at Czech Republic, and analyzed.

## Taxonomy

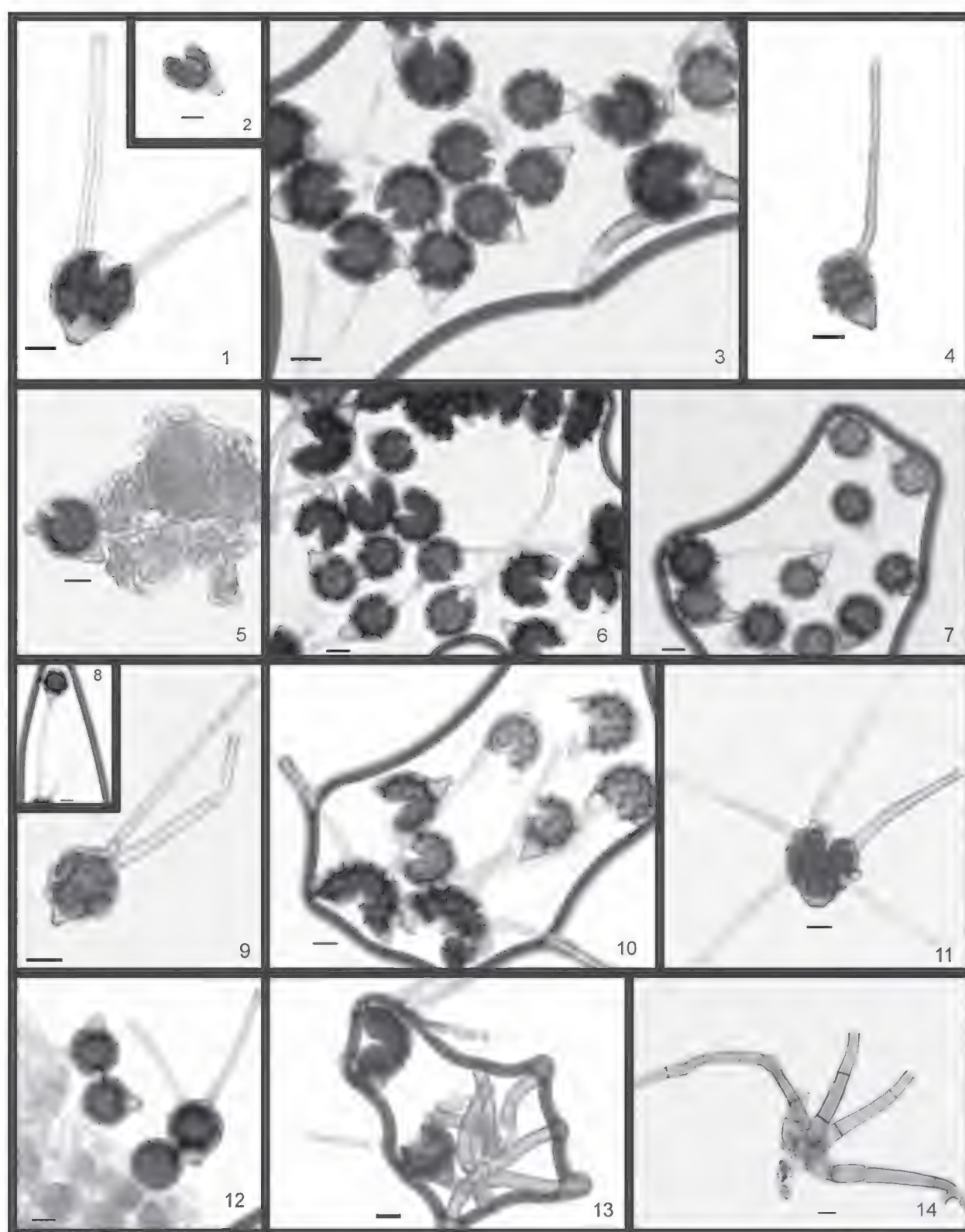
*Chlamydopsis* Hol.-Jech. & R.F. Castañeda, Česká Mykologie 40: 74. 1986.

EMENDED DESCRIPTION: Conidiophores smooth, single or in groups, arising from basal cell. Conidiogenous cell cylindric, smooth, pale brown. Conidium complex, muriform, dry; basal cell obpyriform to subconical, conical-truncate, thick-walled at the base, smooth, pale brown; apical cell globose, dark brown, surrounded by a layer of small, thin-walled, smooth, pale brown cells.

*Chlamydopsis proliferans* Hol.-Jech. & R.F. Castañeda,  
Česká Mykologie 40: 74. 1986.

FIGS 1–14

EMENDED DESCRIPTION: Conidiophores arising from a less distinct basal cell and in groups of up to 5; distinct, simple, 2–7-septate, smooth, pale brown to brown 46–55(–122) µm long, 5–6(–7.5) µm wide, measurement including conidiogenous cells. Conidiogenous cells cylindrical, integrated, terminal, monoblastic, smooth, pale brown, bearing one conidium at the apex. Conidia complex, muriform, solitary, dry, obovoid or obpyriform, with a unicellular basal cell and another terminal portion larger, globose, smooth, brown. Basal cell obpyriform, conical-truncate, thick-walled at the base, smooth, pale brown, 6–8.5 µm long, 6–10 µm wide in the apex, 1–5(–6) µm wide in the base. Terminal portion globose, with a dark brown thick-walled central cell and with a layer of cells covered this portion, 12.5–21 µm diam. Layer of cells surrounding the central cell composing by thin-walled, smooth, pale brown cells, 2–3.5(–5) µm wide. A group of 3–5 conidiophores arising from this layer of cells, simple, 1–3-septate, thin-walled, smooth, pale brown, 37.5–47.5 µm long, 2.5–3.5 µm wide.



FIGS. 1–14. *Chlamydopsis proliferans*. 1–4. Conidia (note thick-walled basal cell). 5–7, 10. Conidia, each with a layer of outer thin cells surrounding the globose dark central cell. 8. Attached conidium. 9–12. Conidiophores arising from the thin outer layer of cells. 13–14. Conidiophores arising from somatic hyphae in groups up to five.

(Bars = 10 µm; FIGS. 1, 2, 4, 5, 9, 11: Brazilian material, SP 381595; FIGS. 3, 6, 7, 8, 10, 12, 13: Cuban isotype, PRM 842703)

SPECIMENS EXAMINED: **BRAZIL. SÃO PAULO:** MOGI-GUAÇU, “RESERVA BIOLÓGICA DE MOGI-GUAÇU”, on decaying leaf litter of *Caesalpinia echinata* Lam. (*Caesalpinaceae*), 30.XII.2005, R.A.P. Grandi & P. Silva. (SP 381595). **CUBA. PROVÍNCIA CAMAGÜEY:** HOYO DE BONET, on rotten leaves of *Lauraceae*, 29.XI.1984, R.F. Castañeda. (ISOTYPE: PRM 842703).

**HABITAT AND DISTRIBUTION** – on leaf litter from tropical rainforest in Brazil and Cuba.

**COMMENTS** – The species was studied through permanent slides from both the Brazilian material and the isotype. In the generic diagnosis the conidia were originally described as uniseptate, with the two cells described as: “terminal cell globose, dark brown, thick-walled and distinctly warted, basal cell subconic, smaller, pale brown, smooth” (Holubová-Jechová & Castañeda Ruiz 1986). However, examination of both collections showed that the conidia are neither warted nor subdivided into two cells. The basal cell of the conidium is conico-truncate at the base as originally described and illustrated and it is thick-walled at the base (FIGS. 1–4). After detailed observations we noted that the “warted” ornamentation of the wall mentioned for the “terminal cell” of the conidia in the original description is actually a lighter coloured layer of cells surrounding the globose dark brown central cell of the conidia (FIGS. 5–7); this species does not have warts. It is well observed that when the conidia are broken, the wall cracks in many directions and the superficial delicate layer is perfectly visible (Fig 1, 2, 5, 6, 10, 11,13). At first the central brown part of the conidia seems to be divided into many cells, but this appearance results from the delicate layer over the globose central cell (FIGS. 5–7, 10). Some cells of this external layer give rise to new conidiophores (FIGS. 8–12); it appears that the conidia may or may not proliferate, depending on the stage of development of the material.

The illustrations in the original paper showed probably 5 conidiophores, which we also observed (FIGS. 13–14), but the species description cites only “up to 4”. In addition, there are no minutely roughened conidiophores observed in the Brazilian material. Unfortunately the illustrations of Holubová-Jechová & Castañeda Ruiz (1986) were at odds with the interpretation in the text.

*Chlamydopsis proliferans* is known only from permanent slides and at the moment its distribution appears to be essentially tropical. This is the second occurrence of the species and the first in South America.

### Acknowledgments

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## MYCOTAXON

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***Amparoina spinosissima*: a continental Asian record  
and some taxonomic observations**

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**Abstract** — *Amparoina spinosissima* is described and illustrated from Kerala State, India. This is the first record of the species from continental Asia. Basidiospores of the Indian specimens are inamyloid in support of Singer's original observation.

**Key words** — *Agaricales*, *Basidiomycota*, floristics, systematics, *Tricholomataceae*

**Introduction**

The genus *Amparoina* Singer (*Agaricales*, *Tricholomataceae*), although little known, has a chequered taxonomic history. The type species of the genus, *A. spinosissima*, was originally described as *Marasmius spinosissimus* and was first discovered in Argentina (Singer 1950). Singer (1958) erected *Amparoina* to accommodate *M. spinosissimus*, which he (Singer 1958, 1976) interpreted as having inamyloid spores, an epicutis covered by cherocytes (loose, globose cells with long excrescences or spines; Singer 1986), and a secotioid habit. Later, Singer (1976) proposed a monotypic family, *Amparoinaceae* Singer, and excluded it from *Agaricales*. Singer (1976) also added a second species to *Amparoina*, *A. heteracantha* Singer. Horak (1980), based on his own collections of *A. spinosissima* made in Argentina and New Caledonia, concluded that the species is not secotioid. After examining the type material of *A. heteracantha*, Horak (1980) considered it to be conspecific with *A. spinosissima*. Horak (1968, 1980), however, never questioned the autonomy of *Amparoina*. Although Singer (1983) did not agree with Horak's merging of the two *Amparoina* species, he conceded that *A. spinosissima* was not secotioid. On the basis of Horak's observations, Singer (1986) reinstated *Amparoina* in the *Tricholomataceae* (*Agaricales*).

Based on the study of several collections of *A. spinosissima* made from Colombia, Puerto Rico, and Hawaii, Desjardin (1995) agreed with most of

Horak's conclusions. However, he found the basidiospores to be amyloid in the specimens he examined and this prompted him to transfer the species to *Mycena* sect. *Sacchariferae*. Although Singer observed the basidiospores of *A. spinosissima* to be inamyloid, this cannot be confirmed as the holotype of *A. spinosissima* no longer exists. Horak's observations (1968, 1980, and his pers. comm. quoted by Desjardin 1995) on the amyloidy of basidiospores from his collections of *A. spinosissima* were not consistent. Takahashi (1999) observed amyloid spores in Japanese collections of the species. We did not re-examine spores from the collections made by Horak and Takahashi, so that the possible variation in amyloid reaction remains an open question. Meanwhile, taxonomic and nomenclatural resources such as the Dictionary of the Fungi (Kirk et al. 2008) and the Index Fungorum ([www.indexfungorum.org](http://www.indexfungorum.org)) continue to recognize *Amparoina*. We accept this point of view for the time being and note that molecular analyses may clarify the relationships among *Amparoina*, *Mycena*, and other agarics in the future. *Mycena* in the present wide sense includes also some species with inamyloid spores as well as species with cherocytes, similar to those of *Amparoina*, on the pileus surface but with amyloid spores (Singer 1986).

Although only rarely collected, *A. spinosissima* is known thus far from Argentina, Colombia, Hawaii, Japan, New Caledonia, and Puerto Rico. During our studies on the agarics of Kerala State, India, we collected a decaying twig bearing primordia of this species, which when incubated in the lab yielded well-developed basidiomata. We present here a full description of the Indian collection along with some taxonomic observations.

### Materials and methods

Conventional morphology-based methods were employed for this study. Microscopic observations were made on material stained with 1% aqueous solutions of phloxine and Congo red and mounted in 3% aqueous KOH. Melzer's reagent was used to observe whether the spores and tissues were amyloid. For statistical evaluation 40 spores (20 basidiospores each from two specimens) were measured. The examined collection cited is deposited at the Kew (Mycology) Herbarium.

### Taxonomy

***Amparoina spinosissima*** (Singer) Singer, Mycologia 50: 110. 1958.      FIGURE 1A–E  
     = *Marasmius spinosissimus* Singer, Schweiz. Z. Pilzk. 28: 193. 1950.  
     = *Mycena spinosissima* (Singer) Desjardin, Bibliotheca Mycol. 159: 15. 1995.

**BASIDIOMATA** small, delicate. **PILEUS** 2–5.5 mm wide, 2–4.5 mm high, initially conical, becoming broadly campanulate; surface white to whitish all over,

entirely covered in the primordial stage with a universal veil made up of pale greenish or ivory-colored, erect or curved, conic, detersile spines up to 0.75 mm long that disappear first from the middle, then from the margin and finally from the pileus disc with age, pruinose, dry, very thin, translucently striate, becoming slightly plicate towards the margin; margin initially straight and appendiculate with spines, becoming plane and undulate or finely torn with age. LAMELLAE adnexed, fairly close, 15–20 reaching the stipe, with lamellulae in 1–3 tiers, ventricose, up to 0.5 mm broad, white; edge finely torn under a lens. STIPE 20–38 × 0.5–1.25 mm, central, terete or slightly compressed, almost equal or with a slightly dilated apex, hollow; surface translucent–white, dry, densely pruinose to hirsute towards the base, almost glabrous at apex; base often subbulbous, not discoid. CONTEXT very thin. ODOR not distinctive.

BASIDIOSPORES (6–)8–9.5(–12) × 4.5–6(–9.5) ( $8.86 \pm 0.17 \times 5.96 \pm 0.15$ )  $\mu\text{m}$ ,  $Q = 1.24\text{--}1.73$ ,  $Q_m = 1.5$ , ellipsoid, ovoid or rarely subamygdaliform, thin-walled, smooth, with refractive guttules, inamyloid. BASIDIA 11–18 × 6–11.5  $\mu\text{m}$ , broadly clavate to almost subglobose, thin-walled, hyaline, 4-spored; sterigmata up to 4  $\mu\text{m}$  long. LAMELLA-EDGE sterile. CHEILOCYSTIDIA 7–23.5 × 5–12.5  $\mu\text{m}$ , cylindrico-clavate, subglobose or vesiculose, covered entirely or at least at the apex with minute excrescences, occasionally smooth, thin- to slightly thick-walled (0.5  $\mu\text{m}$ ), hyaline; excrescences 0.5–0.75  $\mu\text{m}$  long, cylindrical or subconical. PLEUROCYSTIDIA absent. LAMELLAR TRAMA subregular to almost regular; hyphae 2.5–32  $\mu\text{m}$  wide, thin-walled, hyaline to pale yellowish, faintly dextrinoid. PILEAL TRAMA subregular; hyphae 2–20  $\mu\text{m}$  wide, slightly inflated, thin-walled, hyaline to pale yellowish. PILEIPELLIS basically a cutis composed of hyphae that are covered entirely with minute excrescences and terminating in acanthocytes which overlap in such a way as to give an apparent subhymeniform appearance; hyphae 2.5–5.5  $\mu\text{m}$  wide, thin-walled, hyaline; acanthocytes 18–54 × 10–41  $\mu\text{m}$ , versiform: globose, subglobose, clavate, ovoid or sphaeropedunculate, thin-walled, hyaline; excrescences 0.5–2 × 0.5–1.5  $\mu\text{m}$ , cylindrical or subconical; hypoderm composed of distinctly more inflated hyphae lacking excrescences. PILEUS MARGIN made up entirely of cells similar to cheilocystidia, 10–26 × 4.5–15.5  $\mu\text{m}$ , thin-walled, hyaline. SPINES of the universal veil made of cherocytes 25–90 × 2–31  $\mu\text{m}$ , central and terminal ones mostly globose, clavate or fusiform, peripheral ones often cylindric, subcylindric or irregularly elongated, thick-walled (1–2  $\mu\text{m}$ ), with sparse excrescences, with 8–24 erect, pointed spine-like projections, 3–26  $\mu\text{m}$  long. STIPITPELLIS a cutis with numerous caulocystidia; hyphae 2.5–13  $\mu\text{m}$  wide, thin- to slightly thick-walled (0.25  $\mu\text{m}$ ), hyaline; caulocystidia 34.5–331.5+ × 6.5–15(–20)  $\mu\text{m}$ , long, scattered or in clusters, cylindrical, mostly with an obtuse apex, densely covered with excrescences all over. Both acanthocytes and cherocytes observed in the covering layers of the extreme base of the stipe; acanthocytes 11.5–71



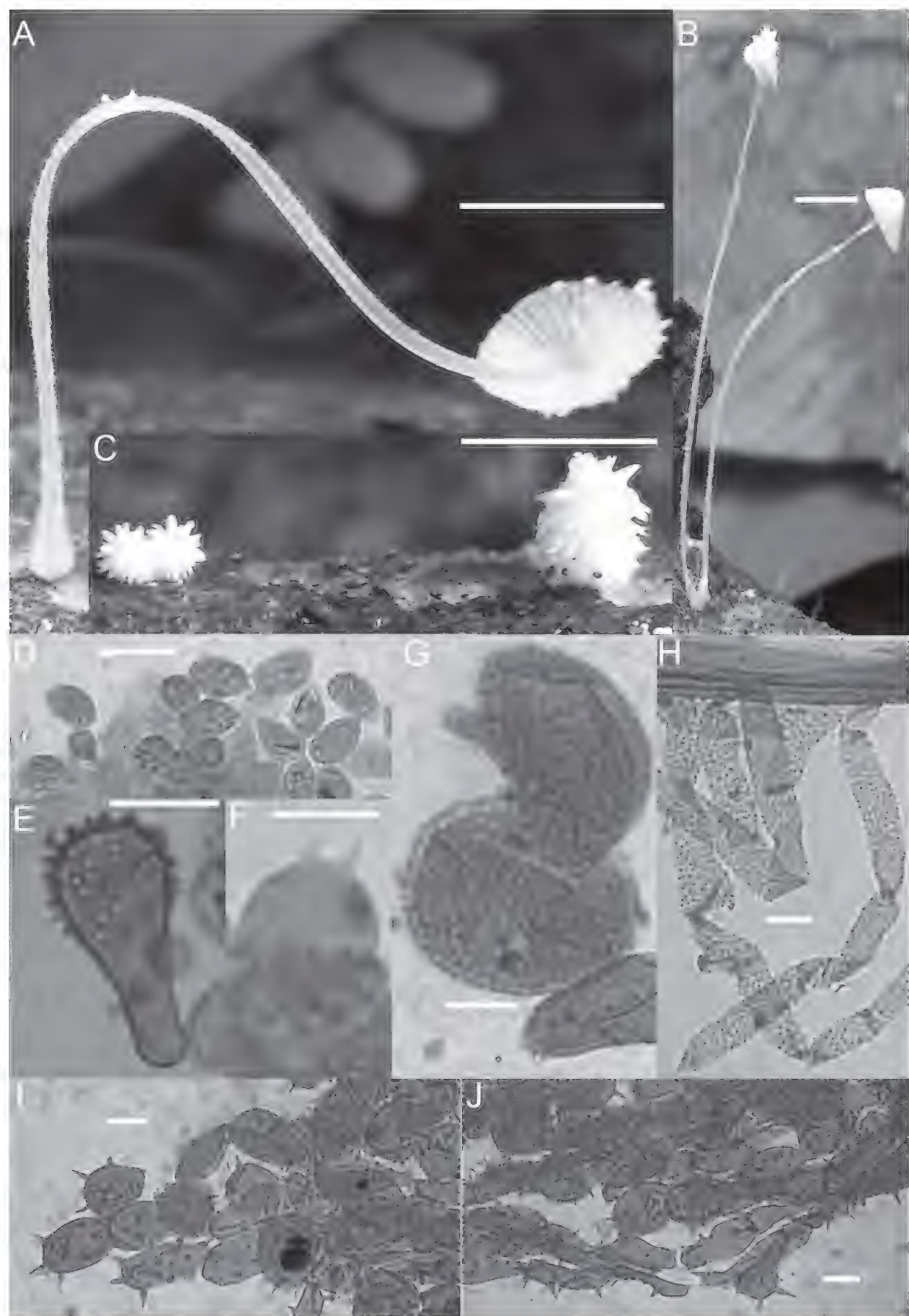


FIGURE 1, A–J: *Amparoina spinosissima*: A–B, basidiomata; C, primordia; D, spores; E, cheilocystidium; F, basidium; G, acanthocytes; H, stipitipellis and caulocystidia; I, cherocytes of terminal part of spine; J, cherocytes of basal part of spine. Scale bars: 5 mm for basidiomata and primordia and 10  $\mu$ m for microscopic structures.



× 7.5–30.5 µm, subglobose to clavate, obpyriform or lageniform or nearly sphaeropedunculate, with evenly distributed excrescences 0.5–2 × 0.5–1.5 µm; cherocytes 23.5–33 × 12–27 µm, globose to subglobose or clavate, thick (1–2 µm)-walled, with excrescences all over, and with 5–12 pointed spine-like projections, up to 8 µm long. CLAMP CONNECTIONS observed in all hyphae except at the base of caulocystidia and on pileipellis hyphae. Cherocytes of both the pileal surface and stipe base showed a tendency to germinate when mounted in water.

HABITAT: On a decaying dicotyledonous twig, scattered or caespitose.

COLLECTION EXAMINED—INDIA, KERALA STATE, Calicut District, KOYILANDY, Poyilkaavu: 31 July 2009, D.M. Aravindakshan DM314 [K(M)165810].

DISCUSSION: The Indian collection shows all diagnostic characters of *A. spinosissima*, such as small, fragile, whitish basidiomata growing on dicotyledonous twigs, a universal veil composed of conical spines comprising thick-walled cherocytes, a pileipellis with deterrent acanthocytes, a stipitipellis with very long and cylindrical caulocystidia with excrescences, cheilocystidia with excrescences, and non-discoid stipe base. However, some minor differences were noticed in the present collection compared to earlier descriptions. In their respective collections, Horak (1980) found all hyphae to be clampless and Desjardin (1995) found clamp connections only in the universal veil. On the contrary, we found clamp connections in most parts of the basidiomata of the Indian collections. While Desjardin found the cherocytes of the medullary region of the spines devoid of spine-like projections, all cherocytes had such projections in the present collections. Additionally, the maximum length of the cherocytes (90 µm), the maximum number of spine-like projections on the cherocytes (24), and the maximum length of the spine-like projections (26 µm) in the Indian collection were almost twice as much as what Desjardin (1995) has recorded. Also, in addition to the normal warty cheilocystidia, occasionally some totally smooth ones were seen. In view of these differences and the reported amyloid spores, Desjardin's collection may represent a different taxon.

As already mentioned, the reaction of the spores of *A. spinosissima* with Melzer's reagent has been a contentious issue and has a bearing on the autonomy of *Amparoina*. The spores of the Indian collection were found beyond any doubt to be inamyloid. This observation lends support for what Singer (1950, 1958, 1976, 1983) has recorded for the species and also for the autonomy of the genus. Another remarkable observation that we made on the Indian specimen is that the cherocytes from the veil tend to germinate when mounted in water. According to Singer (1983, 1986), the cherocytes of *Mycena* and *Amparoina* may be interpreted as chlamydospores.

This is the first record of *A. spinosissima* from continental Asia and it extends the known geographical distribution of this species beyond the Pacific Rim to

South Asia. Our findings support Singer's (1983) contention that *A. spinosissima* has a disjunct distribution and this may be indicative of its primitiveness.

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## MYCOTAXON

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***Hyphopolynema ingae* sp. nov.,  
associated with leaf-spot disease on *Inga edulis* in Brazil**

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**Abstract** — A leaf-spot forming anamorphic fungus, *Hyphopolynema ingae* sp. nov., collected on *Inga edulis* in a fragment of Atlantic forest in Brazil, is described, illustrated and compared with five previously described *Hyphopolynema* species.

**Key words** — appendages, biodiversity, foliicolous fungi, hyphomycetes, taxonomy

## Introduction

*Inga edulis* Mart. (*Mimosaceae*) is a widespread tree in the tropical secondary forest of the Amazonian region and the fragments of Brazilian Atlantic forest (Marangon et al. 2003, Lorenzi 2009). The plant is known by the local population for its sweet edible fruits and antioxidant property of leaves and in folk medicine for its anti-inflammatory and anti-diarrheic properties (Silva et al. 2007, Souza et al. 2007, Lorenzi 2009). During a mycofloristic survey in a fragment of Atlantic forest in the municipality area of Viçosa, Minas Gerais, Brazil, leaves of *I. edulis* showing a leaf-spot disease were collected. On microscopic examination, it was observed that an appendage-bearing anamorphic fungus was associated with the leaf spots. The fungus, which was found to represent a new species of the genus *Hyphopolynema* Nag Raj, is described, illustrated, and discussed in this paper.

## Material and methods

Samples of infected leaves were collected, photographed, and dried in a plant press. Freshly collected samples were examined under a stereomicroscope (Olympus SZ40). Hand sections and fungal material scraped with a scalpel from the plant surfaces were mounted on glass slides with lactophenol. Measurement

and illustrations were carried out with a Carl Zeiss Standard W fitted with a camera lucida drawing apparatus. Photomicrographs were taken in an Olympus BX51 light microscope fitted with a digital camera (Evolt E330). Specimen of the fungus examined was deposited in the Herbarium at the Universidade Federal de Viçosa (Herbarium VIC).

## Taxonomy

*Hyphopolynema ingae* Pinho & O.L. Pereira, sp. nov.

FIGS 1-2

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*Ad Hyphopolynema tropicale differt in cellulae conidiogena 21–33 × 2–5 µm, collis notatis absentibus, setae sporodochio, conidia non guttulata, 0-septata, appendicibus non ramosis.*

**HOLOTYPE:** on leaves of *Inga edulis* Mart. (Mimosaceae), Brazil, Minas Gerais, Viçosa, Reserva Florestal Mata do Paraíso, 6 February 2009, O.L. Pereira (VIC 31222).

**ETYMOLOGY:** from the host genus *Inga*.

Lesions on living leaves, amphigenous, irregular, 0.2–1.4 cm diam., light brown, whitish to grayish at center, surrounded by a purple well defined border, coalescent and necrotic with age. Conidiomata scattered, discrete or often confluent, circular to oval in outline, sporodochial, pulvinate, superficial. Setae sparse in sporodochia, peripheral, erect, straight or slightly curved, medium brown, smooth, 6–9 septate, slightly tapered and paler towards the obtuse apex, 102.5–145.0 × 4.0–5.0 µm. Conidiophores generally reduced to conidiogenous cells, 1–3 septate, pale brown, smooth. Conidiogenous cells terminal, determinate, clustered, integrated or discrete on conidiophores, branched especially at the base, monophialidic, pale brown, smooth, cylindrical or long lageniform and tapered gradually towards the apex, mostly straight, 21.5–37.0 × 2.0–5.0 µm, conidiogenous locus apical, single to each cell, phialide aperture 1.0–2.0 µm wide, with an inconspicuous collarete. Conidia formed in white masses, blastic-phialidic, hyaline, aseptate, smooth, not guttulate, straight, curved or irregular, fusiform, apex acute or rounded, base truncate, often protuberant, 9.0–15.0 × 3.0–6.0 µm; with one apical and 2–4 basal unbranched filamentous appendages, 5.0–10.0 µm long.

**COMMENTS** — Five species have previously been described in the genus *Hyphopolynema*. *Hyphopolynema ingae* is the second species reported on *Mimosaceae*. The other species, *H. tropicale* Nag Raj, is distinguished from *H. ingae* by smaller conidiogenous cells, absence of collarete, absence of setae on conidiomata, guttulate septate conidia, and branched appendages (TABLE 1). *Hyphopolynema tropicale* occurs on pods of *Inga spectabilis* (Vahl) Willd. (Nag Raj 1977), whereas *H. ingae* was found growing on living leaves of *I. edulis*. Among the six *Hyphopolynema* spp., only *H. ingae* and *H. australe* B. Sutton &

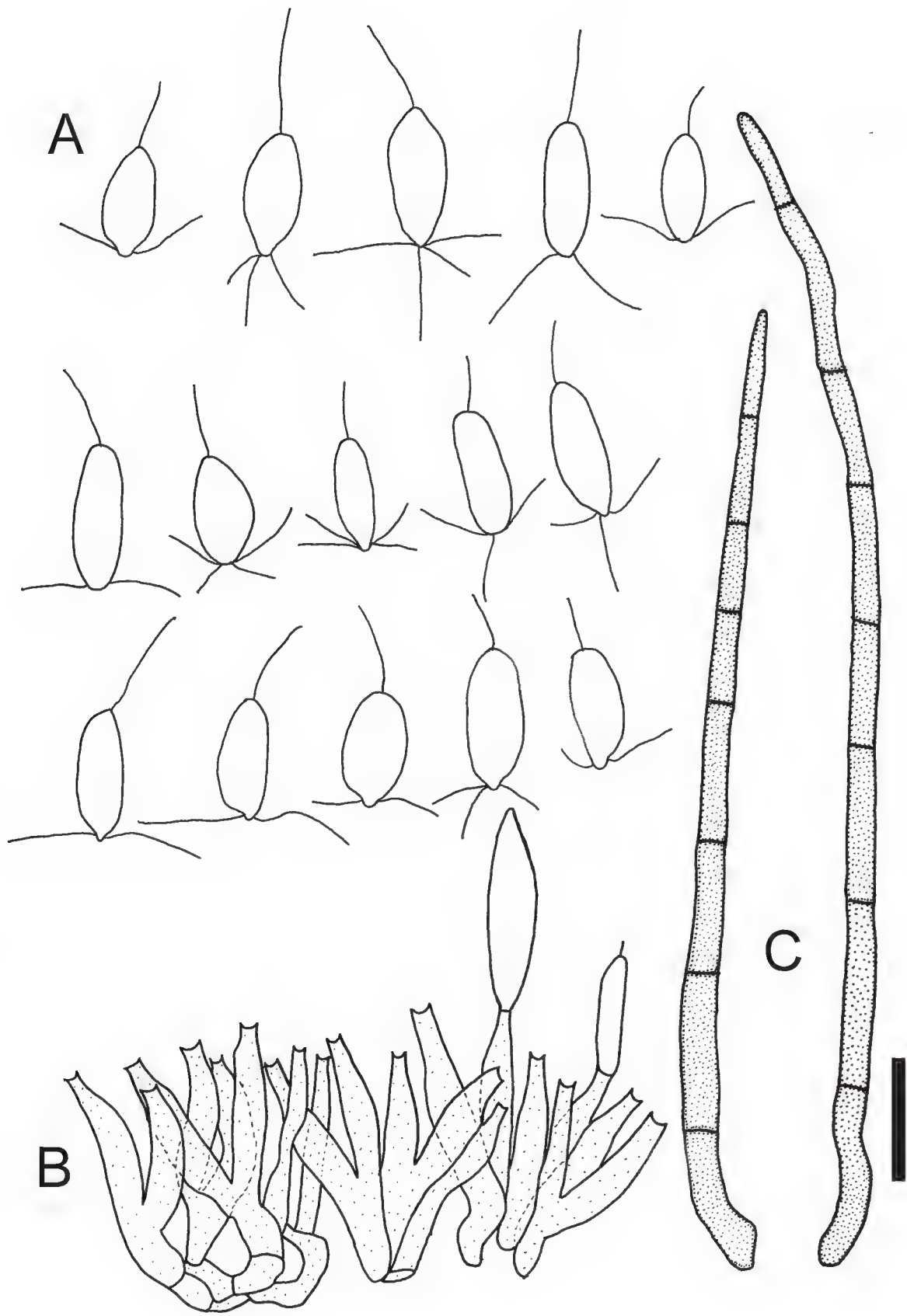


FIGURE 1. *Hyphopolynema ingae* (VIC 31222, holotype) on *Inga edulis*.  
Conidia with flexuous appendages (A).  
Conidiogenous cells arranged in sporodochia (B) and sporodochial setae (C).  
Scale bar = 15 µm.



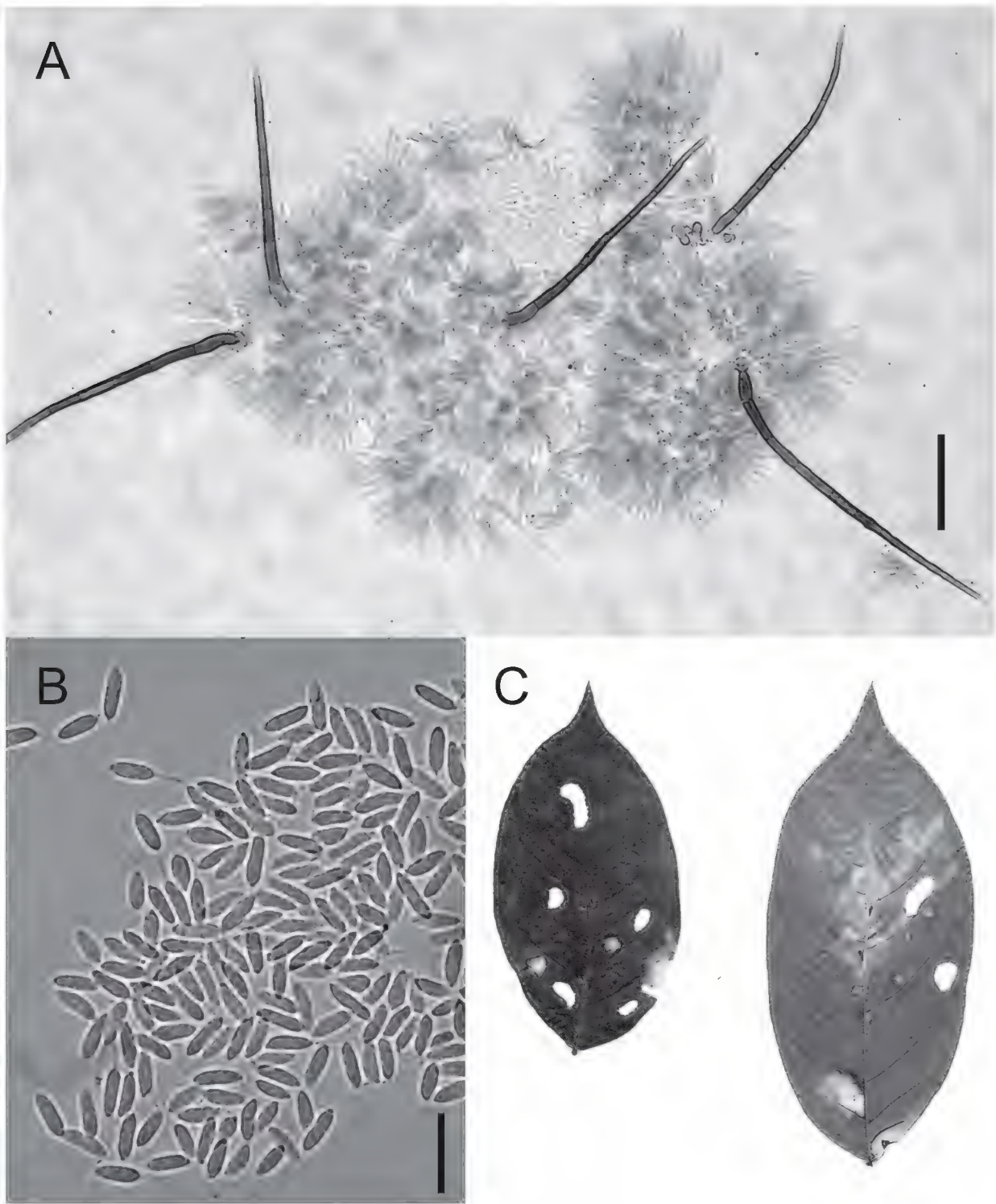


FIGURE 2. *Hyphopolynema ingae* (VIC 31222, holotype). A. Conidiogenous cells arranged in sporodochia. B. Mass of conidia with flexuous appendages. C. Leaf spots associated with *Hyphopolynema ingae* in adaxial and abaxial surfaces from *Inga edulis*.

Scale bars = 40  $\mu\text{m}$  (A); 25  $\mu\text{m}$  (B).

Alcorn are known to occur on living host leaves. In addition, in *H. australe* and *H. ellisiorum* B. Sutton & Alcorn, the conidiophores and conidiogenous cells are hyaline (Sutton & Alcorn 1984). *Hyphopolynema juncatile* Kohlm. & Volkm.-Kohlm. forms a pseudostroma in the cortical tissue of the host (Kohlmeyer

& Volkmann-Kohlmeyer 1999). The sixth species, *H. stilboideum* Bhat & W.B. Kendr., has synnematal conidiomata without setae and produces conidia that are slightly constricted at the septum (Bhat & Kendrick 1993).

TABLE 1. Biometric data (µm) of the species of *Hyphopolynema*.

SPECIES	CONIDIOGENOUS CELLS	CONIDIA	APPENDAGES	SETAE
<i>H. tropicale</i>	11–25 × 3–4	10–17.5 × 4–6	4–9	absent
<i>H. ellisiorum</i>	4–15 × 2.5–4	12.5–13.5 × 2.5–3	7–11	150 × 4
<i>H. australe</i>	7–19 × 2–2.5	15–24 × 2–2.5	4–18	265 × 5–6
<i>H. stilboideum</i>	30–40 × 3–4.5	13–19 × 5–7	8–15	absent
<i>H. juncatile</i>	–	13–16 × 3–4	7–10	55–90 × 4–7
<i>H. ingae</i>	21.5–33 × 2–5	9–15 × 3–6	5–10	102.5–145 × 4–5

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This work is part of an ongoing program of surveying and describing the foliicolous and phytopathogenic mycodiversity of fragments of Brazilian Atlantic forest. The authors wish to thank Prof. Rafael F. Castañeda Ruiz (Instituto de Investigações Fundamentais em Agricultura Tropical “Alejandro de Humboldt”, Cuba) and Prof. Darbhe Jayarama Bhat (Goa University, India) for reviewing the manuscript.

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## MYCOTAXON

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**A new species of *Entoloma* from Western Ghats of India**

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**Abstract** — A new species, *Entoloma vittalii* (sect. *Cyanula*, subg. *Leptonia*, *Entolomataceae*), collected from paleotropical regions of the Uppangala forest, Western Ghats, Karnataka, is described and illustrated. Macro- and microscopic differences and similarities are compared with closely related taxa.

**Key words** — *Agaricales*, *Basidiomycota*, fungal taxonomy, macrofungi

**Introduction**

Species of *Entoloma*, one of the largest genera in the *Agaricales*, are distributed throughout the world. In India, Pegler (1977) revised descriptions of *Entolomataceae* species, and Horak (1980) also treated several entolomatoid taxa. Manimohan et al. (1995, 2002, 2006) contributed the most notable records, describing 39 *Entoloma* species from Kerala state alone. As a result, a total of 69 entolomatoid species have been described from different regions in India (Manjula 1983, Natarajan et al. 2005). During our studies on diversity of macrofungi from Western Ghats of Karnataka, we collected several *Entoloma* species, of which six represented first records for India (Senthilarasu & Natarajan 2003). One species, which differs macro- and microscopically from known allied species, is described below as new to science.

**Materials and methods**

The collections described here are from paleotropical regions of the Uppangala forest, Western Ghats, Karnataka, India. Sections were prepared by hand, revived

in 10% KOH and examined in 2% phloxine. Approximately 50 basidiospores obtained from spore prints were measured. Mean spore measurements (in parentheses) are followed by spore size range, with extreme values in parentheses. Colour terminologies follow Kornerup & Wanscher (1978). The examined type specimens are deposited at Herbarium of Madras University Botany Laboratory (MUBL).

***Entoloma vittalii* Senthil., Kumaresan & S.K. Singh, sp. nov.**

PLATE 1

MYCOBANK MB 518331

*Pileus* 35–45 mm *latus*, *plano-convexus*, *umbonatus*, *acutus ad discum rubrobrunneus*, *ad marginem fuscus*, *laevis*, *glaber*; *marginem laevis*, *erosus*, *lucido-striatus*. *Lamellae emarginatae*, *cremeus*, *pallens aurantiacus*, *confertae*, *latissimae*, *cum tribus ordinibus lamellarum intermixtae*. *Stipes* 40–55 × 6–10 mm, *cylindricus vel expansus*, *cavus*, *ad apicem lilacinus*, *ad basim lilacinus griseus*, *laevis*, *glaber*. *Caro tenuissima*, *albida*, 3 mm *latus*. *Sporae* (8.9 ± 0.6 × 6.1 ± 0.4), (7–)7.5–10(–10.5) × 5.5–7(–7.5) µm, Q = 1.45, *heterodiametrico-ellipsoidea*, *angulatae*. *Basidia* 24–34 × 7.5–9.5 µm, *clavata*, 4 *sporigera*. *Acies lamellarum fertilis*. *Cystidia nulla*. *Trama hymenophoralis regularis*, *hyalina*. *Epicutis ex hyphis cylindricus*, 1.5–12 µm *latus*. *Hyphae omnes defibulatae*.

HOLOTYPE: India, Karnataka State, Western Ghats, Manadukka, Uppangala Forest, 12°30'N 79°30'W, 500 masl, on ground (soil), Senthilarasu G. (MUBL 3496).

ETYMOLOGY: This species is named in honor of Prof. B.P.R. Vittal of the Centre for Advanced Studies in Botany, University of Madras, India.

*Pileus* 35–45 mm diam., *plano-convex*, becoming *uplifted*, *acutely umbonate*; surface reddish brown (8F8) at the center, paler (8D4) towards margin, smooth, glabrous; margin smooth, eroded, pellucid striate. *Lamellae emarginate*, cream, becoming pale orange (5A3), crowded, moderately broad with lamellulae of three lengths. *Stipe* 40–55 × 6–10 mm, *cylindric to compressed*, hollow; surface violet white (15A2) at the apex, lilac grey (15B2) below, smooth, glabrous, arising from white, rhizomorphs. Context thin, whitish, up to 3 mm thick.

Basidiospores (8.9 ± 0.6 × 6.1 ± 0.4), (7–)7.5–10(–10.5) × 5.5–7(–7.5) µm, Q = 1.45, heterodiametric-elliptic, with well marked angles, with 5–7 occasionally 8 plane to few concave facets visible in profile, with a thickened stramineous wall, containing a single, large refractive guttule. Basidia 24–34 × 7.5–9.5 µm, clavate, bearing four sterigmata, up to 5.5 µm long. Lamella-edge fertile. Cystidia absent. Hymenophoral trama regular, with hyaline, thin-walled hyphae, 1.5–11.5 µm diam. inflated to 17 µm diam. Subhymenial layer poorly developed, up to 6 µm wide, interwoven. Pileal surface a repent epicutis of radially arranged parallel hyphae, 1.5–12 µm diam. Pileal context consisting of tightly interwoven, thin-walled, hyaline hyphae, 1.5–17.5 µm diam., inflated to 37.5 µm diam. All hyphae lacking clamp-connections.

HABITAT - On ground, solitary, scattered in wet evergreen forest.





PLATE 1. *Entoloma vittalii*: 1–2—In situ, Uppangala forest (Photo G. Senthilarasu): 1. Habit. 2. Gill view. 3—Line drawings (a–b.  $\times 1$ ; c–d, bar = 10  $\mu\text{m}$ ): a. Habit. b. Gill view. c. Basidiospores. d. Basidia.

DISCUSSION—The characteristic features of *Entoloma vittalii* are the plano-convex to uplifted and acutely umbonate, reddish brown, smooth pileus, violet white to lilac grey stipe, heterodiametric-elliptic spores, and absence of cystidia. Species with uplifted, acutely umbonate reddish brown pilei with violet white to lilac grey stipes are uncommon, and very few species have been reported in the literature. *Entoloma vittalii* seems to fit best in subg. *Leptonia*, sect. *Cyanula* (Noordeloos 1992) based on collybioid habit, violaceous stipe, heterodiametric basidiospores, and lack of cheilocystidia and clamp connections. However, the umbonate, glabrous pileus is somewhat out of place for this subgenus and section, which are typically defined by an umbilicate, squamulose pileus surface. It is not clear at this time where *E. vittalii* belongs in the genus as recognized by Noordeloos (1992).

*Entoloma vittalii* resembles *E. parvum* (Peck) Hesler (Hesler 1967) in similarly sized basidiomes, heterodiametric elliptic spores, and absence of cystidia. However, its conic-convex, bluish black pileus, adnate lamellae, and bluish black stipe clearly differentiate *E. parvum* from *E. vittalii*.

*Entoloma vittalii* also closely resembles the paleotropic species *E. maderaspatanum* (Pegler) E. Horak (Horak 1980) in having an umbonate, brown, smooth pileus and lacking cheilocystidia and clamp-connections. However, *E. maderaspatanum* clearly differs in its conic-convex, dark brown pileus, long (8 cm vs 4–5.5 cm) white or cream colored stipe, and somewhat larger spores (9–12.5  $\mu\text{m}$  vs 7–10.5  $\mu\text{m}$ ).

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## **New records of smut fungi. 2. *Anthracoidea arnellii* sp. nov.**

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**Abstract** — A new smut fungus, *Anthracoidea arnellii* on *Carex arnellii*, is described and illustrated from Russia.

**Key words** — *Anthracoideaceae*, taxonomy, *Ustilaginomycetes*

### **Introduction**

A specimen of *Carex arnellii* from the Altai Mts (West Siberia), Russia was found to be infected by an undescribed species of *Anthracoidea* smut fungus. *Carex arnellii* is a member of the sect. *Silvaticae* Rouy, which includes nine species and subspecies from Europe, Asia, and North Africa. *Carex arnellii* is distributed in the European part of Russia, West and East Siberia, the Russian Far East, northern Mongolia, NE China, and the northern part of the Korean Peninsula (Egorova 1999). The species of *Anthracoidea* are restricted to host plants belonging to the same or closely related sections of *Carex*. No species of *Anthracoidea* has previously been reported on a representative of sect. *Silvaticae*.

### **Material and methods**

Material from the herbarium of Komarov Botanical Institute, Russian Academy of Sciences, St Petersburg (LE) was examined under light microscope (LM) and scanning electron microscope (SEM). For LM observations, the spores were

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mounted in lactophenol solution on glass slides, gently heated to boiling point, and then cooled. The measurements of spores are given in the form: min–max (mean  $\pm$  1 standard deviation). For SEM, the spores were attached to specimen holders by double-sided adhesive tape and coated with gold with an ion sputter. The surface structure of spores was observed at 10 kV and photographed with a JEOL SM-6390 scanning electron microscope.

## Taxonomy

***Anthracoidea arnellii*** Denchev, T. Denchev & Karatygin, **sp. nov.**

FIGS 1–4

MYCOBANK MB 518336

*SORI* in ovarii in inflorescentia dispersi, sicut corpora subglobosa, late ellipsoidea vel ovoidea, nigra, 2–3 mm longa, in superficie pulverei. *SPORAE* irregulariter polyangulares, interdum protuberantibus, a fronte visus  $16.5\text{--}26 \times 14.5\text{--}20.5$  ( $20.4 \pm 1.9 \times 17.9 \pm 1.5$ )  $\mu\text{m}$ , a latere visus  $11.5\text{--}13.5$   $\mu\text{m}$ , rufobrunneae; paries inaequaliter incrassatus,  $1\text{--}2.5$  ( $-3$ )  $\mu\text{m}$  crassus, plerumque  $1\text{--}3$  ( $-4$ ) gibberis internis, et raro etiam maculis lucem refringentibus; superficie verruculosa.

**HOLOTYPE:** On *Carex arnellii* Christ: RUSSIA, Altai Republic, the Altai Mts, near Teletskoe Lake, valley of Chiri River, 3 August 1985, leg. I.V. Karatygin (LE 68 682).

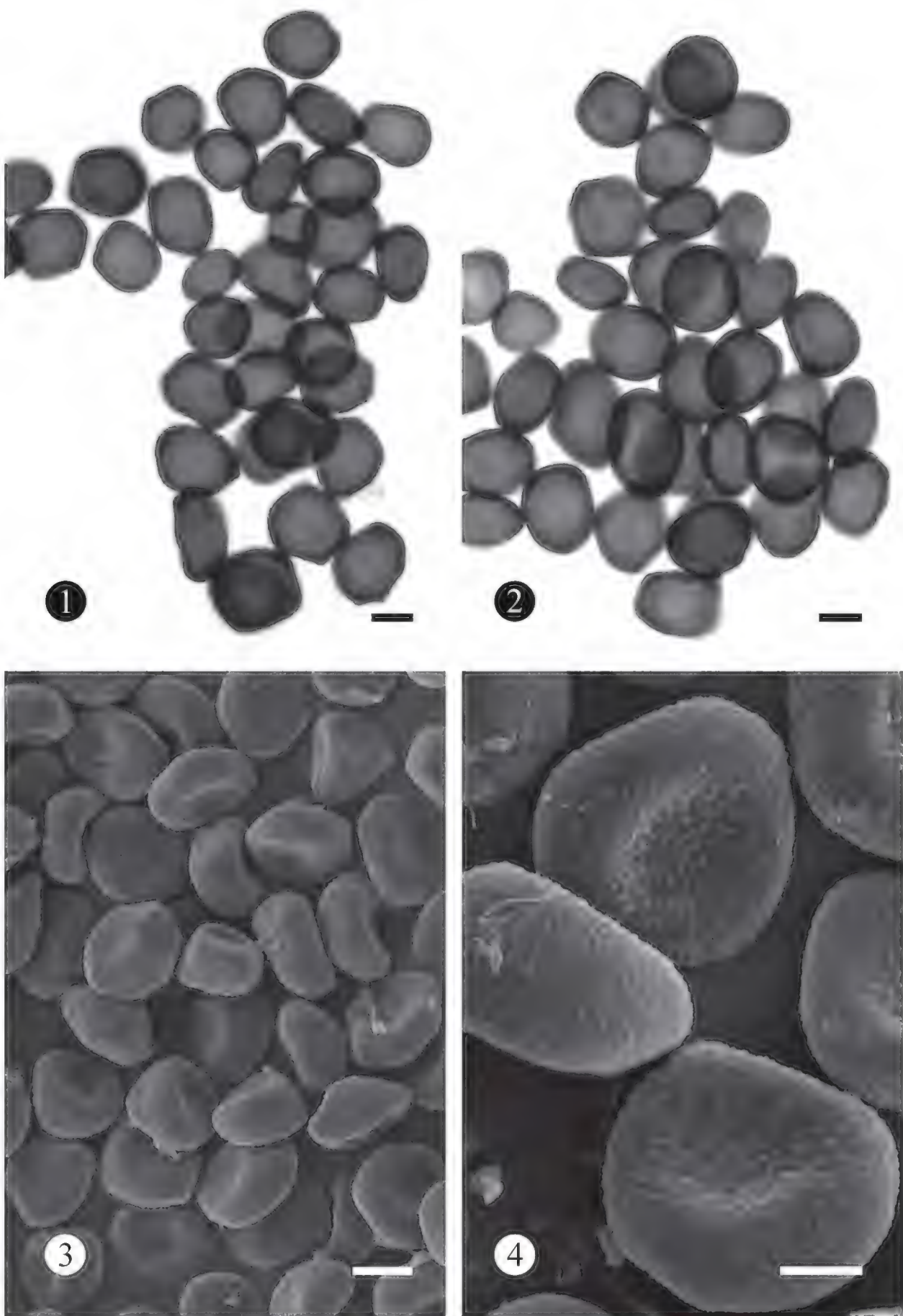
**ETYMOLOGY:** the name refers to the host species.

*SORI* in ovaries, scattered in the inflorescence, as subglobose to broadly ellipsoidal or ovoid, black, hard bodies, 2–3 mm long, when young covered by a thin, silvery membrane; later becoming exposed but partly hidden by the glumes; mature sori powdery on the surface. *SPORES* irregularly polyangular, sometimes with protuberances, in plane view  $16.5\text{--}26 \times 14.5\text{--}20.5$  ( $20.4 \pm 1.9 \times 17.9 \pm 1.5$ )  $\mu\text{m}$  ( $n = 50$ ), in side view  $11.5\text{--}13.5$   $\mu\text{m}$  thick, reddish brown; wall unevenly thickened,  $1\text{--}2.5$  ( $-3$ )  $\mu\text{m}$  thick, thickest at the angles and protuberances, with  $1\text{--}3$  ( $-4$ ), distinct internal swellings, rarely with light-refractive areas, verruculose. Germination unknown.

**DISTRIBUTION** — On *Cyperaceae*: *Carex* (subgen. *Carex*, sect. *Silvaticae*), Asia (West Siberia, the Russian Far East).

**COMMENTS** — On *Carex arnellii*, Kawai & Ôtani (1931: 230) reported *Anthracoidea* sp. (as “*Cintractia caricis*”) from Sakhalin (the Russian Far East; collected on 23 July 1930 by E.C. Higashi-Taraika). Unfortunately, there is no information if this specimen is kept in any Japanese herbarium.

*Anthracoidea arnellii* possesses irregularly polyangular spores with distinct internal swellings like *A. capillaris* Kukkonen but the spores of the latter are smaller. *Anthracoidea capillaris* is known to attack only *Carex capillaris* L. In older taxonomic schemes, *Carex arnellii*, *C. sylvatica* Huds., and *C. capillaris* were included in sect. *Strigosae* Christ (Chater 1980). In recent taxonomic schemes (e.g., in Egorova 1999), the three species are treated as members of two different, non-related sections: *C. arnellii* and *C. sylvatica* in *Silvaticae*,



FIGS 1–4. Spores of *Anthracoidea arnellii* on *Carex arnellii* (holotype).  
1–2. In LM. 3–4. In SEM. Scale bars: 1–3 = 10  $\mu$ m, 4 = 5  $\mu$ m.

and *C. capillaris* in *Chlorostachyae* Meinsh. (synonyms: sect. *Hymenochlaenae* subsect. *Capillares* (Asch. & Graebn.) Kük.; sect. *Capillares* (Asch. & Graebn.) Rouy). For *Carex sylvatica* and *C. capillaris*, Hendrichs et al. (2004) found that they “are neither clustered together nor with any other member of section *Hymenochlaenae*” and that the section *Hymenochlaenae* is heterogeneous. Because of these reasons, we consider *Anthracoidea arnellii*, on a member of sect. *Silvaticae*, as a distinct species.

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***Cetraspora helvetica*, a new ornamented species in the  
*Glomeromycetes* from Swiss agricultural fields**FRITZ OEHL<sup>1</sup>, JAN JANS<sup>2</sup>, FRANCISCO ADRIANO DE SOUZA<sup>3</sup>,  
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**Abstract** — A new arbuscular mycorrhizal fungus, *Cetraspora helvetica*, was found in three Swiss agricultural soils: a no-till crop production system and two temporary grasslands. It forms white spores, 210–270 µm diam, on dark yellow sporogenous cells. The spores have three walls: a triple-layered outer, a bi-layered middle and a triple-layered inner wall. The spore surface is crowded with convex warts, 5–12 µm diam at the base and 1.5–5.0 µm high. The germination shield is hyaline with multiple (6–10) lobes. Glomerospores of two other *Gigasporineae* spp. have also three walls, multiple-lobed hyaline germination shields, and projections on the outer spore surface: *C. spinosissima* and *C. striata*. However, spores of these fungi are substantially pigmented (ochraceous yellow to rust) and crowded with short, thin spines or fingerprint-like processes, respectively. Partial sequences of the 28S ribosomal gene place the new species adjacent to *C. spinosissima*, *C. pellucida*, and *C. gilmorei*. Phylogenetic analyses demonstrate the monophyly of the two genera *Racocetra* and *Cetraspora* within the *Racocetraceae*.

**Key words** — *Gigasporaceae*, *Glomeromycota*, *Scutellospora*, conservation tillage

**Introduction**

Several species of the *Gigasporineae* sensu Morton & Benny (1990) have been recently described (e.g. Silva et al. 2008, Goto et al. 2009, 2010; Tchabi et al. 2009). However, most of the so far nearly 50 species described in this old sub-order have been known only for the warmer climates, and indeed species

richness of the *Gigasporineae* appears to be much lower in colder climates, especially in Europe north of the Alps (e.g. Jansa et al. 2002, Oehl et al. 2009b, 2010). In Northern and Central Europe, so far only ten species of this group has been found: *Gigaspora margarita* W.N. Becker & I.R. Hall 1976, *G. gigantea* (T.H. Nicolson & Gerd.) Gerd. & Trappe 1974, *Scutellospora calospora* (T.H. Nicolson & Gerd.) C. Walker & F.E. Sanders 1986, *S. dipurpurescens* J.B. Morton & Koske 1988, *S. arenicola* Koske & Halvorson 1990, *S. nodosa* Błaszk. 1991, *Racocetra castanea* (C. Walker) Oehl et al. 2009, *R. persica* (Koske & C. Walker) Oehl et al. 2009, *Cetraspora pellucida* (T.H. Nicolson & N.C. Schenck) Oehl et al. 2009, and *C. armeniaca* (Błaszk.) Oehl et al. 2009 (e.g. Błaszkowski 1991, Błaszkowski & Tadych 1997, Tadych & Błaszkowski 2000, Vestberg et al. 1995, Merryweather & Fitter 1998, Jansa et al. 2002, 2003, Oehl et al. 2005, 2010). Our morphological and molecular analyses revealed that one isolate, registered at the International Bank for *Glomeromycota* (BEG) and identified as *Scutellospora pellucida* (= *C. pellucida*) is not *C. pellucida* but a closely related, undescribed species whose spores have conspicuous warty ornamentation on the outer surface. The fungus, collected from three agricultural soils in Switzerland, is here described under the epithet *Cetraspora helvetica*.

## Material and methods

### Study area and sites

Between 1998 and 2009, AMF species richness was determined in > 100 Swiss agricultural soils distributed all over the country and including permanent grassland, conservation and no-tillage, biological and conventional agroecosystems (e.g. Jansa et al. 2002, Oehl et al. 2003, 2004, 2010, Oehl unpublished). The AMF communities of these sites were propagated in bait cultures for 8–32 months, in most of the cases for 18–24 months. At only three sites, the AM fungus, which is hereafter described, was detected. The sites are a long-term tillage experiment at Agroscope ART in Tänikon (Kanton Thurgau, 47°29'10.0"N, 8°55'10.1"E, at 540 m a.s.l.), a temporary grassland in the community Langnau im Emmental (Kanton Bern, 46°56'35.0"N, 7°45'46.8"E, at 656 m a.s.l.), and a temporary grassland in the community Grasswil (Kanton Bern, 47°08'34.0"N, 7°39'58.2"E, at 525 m a.s.l.). The locations have a temperate climate (typical for Central Europe), with mean annual temperatures of 8.5, 8.0, and 9.2 °C and mean annual rainfall of about 1200, 1450, and 1100 mm, respectively.

### Soil sampling and soil parameters

In Tänikon, soils were sampled in January 1999 as described in Jansa et al. (2002). The soil samples in Langnau were sampled accordingly in April 2009. The soil type in Tänikon and Grasswil is a Haplic Luvisol developed on Moräne, while the soil type in Langnau was a Fluvic Cambisol developed on alluvial sediments. The pH (H<sub>2</sub>O) of the topsoil was 6.0 at all three sites. The organic carbon content was 19.1, 21.1 and 12.2 mg C g<sup>-1</sup> soil at Tänikon, Langnau and Grasswil, respectively. Total N and available P (according to Dirks & Scheffer 1930) were 2.3, 2.5 and 3.0 mg N g<sup>-1</sup> soil and 2.3, 2.2, and 7.4 mg P kg<sup>-1</sup> soil, respectively.



### AMF bait and pure cultures

The AMF bait cultures for the soil from Tänikon were established at ETH Zurich in Eschikon-Lindau (Kanton Zurich) as described in Jansa et al. (2002) using *Zea mays* L., *Allium porrum* L., *Plantago lanceolata* L., *Helianthus annuus* L. and *Glycine max* (L.) Merr. as bait plants. The bait cultures for the soils from Langnau and Grasswil were established at Agroscope ART in Zürich-Reckenholz on *P. lanceolata*, *Lolium perenne* L., *Trifolium pratense* L., and *Hieracium pilosella* L. as host plants, as described for *Acaulospora alpina* (Oehl et al. 2006) but with the pots substantially larger than in the former work (volume 3.5 L instead of 1.0 L).

Pure cultures of the new fungus were established by inoculating leek plants with 15 spores obtained from the Tänikon soil bait cultures. The cultures have been maintained for several cycles of 15-24 months at ETH (alternating *A. porrum* and *Tagetes erecta* L. as host plants, and on *Medicago truncatula* L.). The isolate was also deposited in the European Bank of *Glomeromycota* under the accession number BEG153 and is maintained in the Swiss Collection of Arbuscular Mycorrhizal Fungi (SAF) at Agroscope ART in Zürich-Reckenholz under the accession number SAF15.

### Morphological analyses

Glomerospores were extracted from field soils by wet sieving (Gerdemann & Nicolson 1963) and sucrose centrifugation (Jenkins 1964). The spores were thereafter mounted in polyvinyl-alcohol-lacto-glycerin (PVLG), PVLG + Melzer's reagent, and water (Brundrett et al. 1994, Spain 1990). About 100 spores of the fungus were examined. For the species description, terminology follows that used for the *Diversisporales* by Oehl et al. (2006), Sieverding & Oehl (2006), and Palenzuela et al. (2008, 2010), for germ shield structures by Walker & Sanders (1986), Oehl et al. (2009a), and Goto et al. (2010), and for spore denomination by Goto & Maia (2006).

Spore wall composition was compared with that observed in spores in type specimens of *Cetraspora armeniaca* [ex type: Błaszowski collection], *C. gilmorei* (Trappe & Gerd.) Oehl et al. 2009 [Holotype OSC 30'990; paratype OSC 31'018; paratype OSC 30'921, *C. pellucida* [Holotype OSC 37'515], *C. spinosissima* (C. Walker & Cuenca) Oehl et al. 2009 [Ex type: (Gisela Cuenca collection, Oehl collection), and *C. striata* (Cuenca & R.A. Herrera) Oehl et al. 2009 [Ex type: Gisela Cuenca collection, slides 1642-7 & 1641-3].

### Molecular analyses

The DNA from single spores was extracted according to Sanders et al. (1995). Single spores were crushed in 10 µl of PCR-grade water by freshly flamed Pasteur pipette. After 5 µl of Chelex-100 (20%, Bio-Rad Laboratories, Hercules, California, USA) were added, samples were placed onto a 95°C hot plate for 3 minutes and then incubated on ice at 0°C for 5 minutes. Five µl of the liquid phase were taken as template for PCR amplification of the large ribosomal subunit gene, 28S.

Spore DNA samples underwent a nested PCR procedure, first using eukaryotic-specific primers ITS3 and NDL22 (White et al. 1990), followed by fungal-specific primers LR1 and FLR2 (van Tuinen et al. 1998; Turnau et al. 2001). There were 30 cycles with each primer pair. The product of the first PCR was cloned or further diluted 1000 times, and 5 µL of the diluted mixture was used as a template for the second PCR reaction. PCR conditions followed van Tuinen et al. (1998), the annealing temperature

being 60°C in both PCR steps. The PCR products were then purified using QIAquick PCR Purification Kit (Qiagen Sciences), cloned into a blue script vector (pGEM-T Easy, Promega-Catalys AG, Wallisellen, Switzerland), and transformed into bacterial strain *E. coli* JM109 by the heat-shock method. The size of the insert in growing bacterial colonies was checked after PCR amplification using M13f and M13r primers that were targeted to the cloning site of the vector. Plasmid DNA was isolated from transformed bacteria following standard miniprep procedure (Sambrook et al. 1989), and used as a template for cycle sequencing using BigDye Terminator (Applied Biosystems, Foster City, California, USA). Sequencing analysis was performed on ABI-310 Capillary Sequencer (PerkinElmer, Wellesley Massachusetts, USA). Four sequences were obtained and deposited at GenBank (National Center for Biotechnology Information, Bethesda, Maryland, USA) under the accession numbers AF396784 and HM565944–HM565946.

The sequences of the new species were aligned with other glomeromycotean sequences from the GenBank using ClustalX (Larkin et al. 2007) and edited by BioEdit (Hall 1999) to obtain a final alignment.

For phylogenetic analyses and tree construction, maximum parsimony (MP) and neighbor joining (NJ) analyses with 1000 bootstrap replications for each, were performed using the Phylogenetic Analysis Using Parsimony program version 4 (Swofford 2003). The NJ analysis was performed using parameters obtained from ModelTest 3.7 (Posada & Crandall 1998). Sequences from *Pacispora scintillans* were used as outgroup.

## Taxonomy

***Cetraspora helvetica*** Oehl, Jansa, F.A. Souza, G.A. Silva, **sp. nov.**

FIGS. 1–19

MYCOBANK MB 518578

*Sporocarpia ignota*. Sporae singulatim in solo efformatae anguste adiacetae ad cellulas sporogeneas subterminales vel intercalares flavasque, albae, globosae (210–270 µm in diametro) vel subglobosae vel ovaes (205–265 × 210–280 µm); sporae tunicis tribus: tunica exterior stratis tribus, in totum 8.4–15.0 µm crassa; stratum exterius tunicae exterioris hyalinum, semi-persistens, 0.9–1.6 µm crassum, cum verrucis exiguis 1.5–5.0 altis et 5–12 µm latis; stratum medium laminatum, album, 8.4–15.0 µm crassum; stratum interius tunicae exterioris album, 1.0–1.6 µm crassum; tunica media stratis duobus hyalinibus, 1.5–2.5 µm crassa in totum; tunica interior stratis tribus hyalinibus, 2.5–5.9 µm crassa in totum; stratum secundum et stratum interius tunicae exterioris et stratum secundum tunicae interioris purpureum vel oscuro-purpureum colorantes reagente Melzeri; scutellum germinale in superficie exteriore tunicae interioris, hyalinum ad subhyalinum ad albo-flavum; subglobosum vel ovale vel ellipsoidum, 120–150 × 120–200 in diameter, lobatum, paucioribus 6–10 lobis; structurae mycorrhizarum arbuscularum colorantes caeruleae Trypan blue; cellulae auxiliares formans.

TYPE: SWITZERLAND: Kanton Thurgau, Tänikon, Agroscope Reckenholz-Tänikon Research Station (ART), from agricultural soil under no-till wheat–maize–canola production, 1999 by J. Jansa". (**Holotype**: 88-8801 (Z+ZT Myc 3037); pure cultures—ETZ Zürich (Eschikon) and Swiss Collection of Arbuscular Mycorrhizal Fungi (SAF; Zürich Reckenholz). **Isotypes**: 88-8802, 88-8803, 88-8804, 88-8805, 88-8806 (Z+ZT Myc 3038); 88-8807, 88-8808 (OSC 136'595); 88-8809, 88-8810 (URM).

ETYMOLOGY: *helvetica* (Latin) = Swiss, referring to the country where the fungus was detected first.

GLOMEROSPORES formed singly in soil, terminally on subterminal or sometimes intercalary bulbous suspensor cell (= 'sporogenous' cell; FIGS. 1–6). Glomerospores are brilliant white when young, and may slightly darken to creamy-white when ageing in soils, trap culture substrates or after several months in lactic acid based mountants (FIGS. 1–3). The spores are globose (210–270  $\mu\text{m}$  in diameter) to subglobose (205–265  $\times$  210–280  $\mu\text{m}$ ), become dark purple to black purple when exposed to Melzer's reagent (FIG. 5), and have three walls: an outer, a middle, and an inner wall (FIG. 7).

OUTER WALL is 8.4–15.0  $\mu\text{m}$  thick in total and consists of three layers (FIGS. 7–8): outermost wall layer (OWL1) is hyaline, semi-persistent and 1.1–1.6  $\mu\text{m}$  thick crowded with convex warts that are 5–12  $\mu\text{m}$  in diameter at their base and 1.5–5.0  $\mu\text{m}$  high (FIGS. 8–9). OWL2 is brilliant white, and may become creamy-white with age. It is laminate, persistent and 5.0–12.0  $\mu\text{m}$  thick. Third layer (OWL3) is also white, semi-flexible (1.0–1.6  $\mu\text{m}$  thick). OWL2 and OWL3 stain dark purple to black purple in Melzer's reagent, while OWL1 generally does not stain (FIG. 8). The straight pore channel at the spore base (about 2.5–3.9  $\mu\text{m}$  broad) is often closed by a plug formed by spore wall material of OWL2, and by OWL3, but also can appear open.

MIDDLE WALL (MW) is 1.8–2.7  $\mu\text{m}$  thick in total and consists of two hyaline layers: a flexible outer layer MWL1 and a semi-flexible layer MWL2 (FIGS. 7, 10). MWL1 is 0.7–1.2  $\mu\text{m}$  thick and generally does not separate from underlying MWL2 but often shows several folds in crushed spores (FIG. 10). MWL2 is 1.1–2.0  $\mu\text{m}$  thick, and generally more rigid than MWL1.

INNER WALL (IW) is triple-layered (FIGS. 7), 2.5–4.5(–5.9)  $\mu\text{m}$  thick, bearing a germ shield on the outer surface (FIG. 4, 11). The outer IW layer (IWL1) is hyaline, semi-flexible and 0.6–0.8  $\mu\text{m}$  thick. The second layer (IWL2) is semi-flexible, unite to finely laminate, amorphous when slightly expanding in PVLG based mounting, and is 2.0–2.7(–3.9)  $\mu\text{m}$  thick. The innermost layer (IWL3) is relatively thin (0.6–1.2  $\mu\text{m}$  thick), flexible, mostly tightly adherent to IWL2, and therefore generally difficult to observe. IWL2 stains purple to dark purple to black purple in Melzer's reagent (FIG. 11).

SPOROGENOUS CELL (sc) is globose to elongate, 34–70  $\mu\text{m}$  long and 30–48  $\mu\text{m}$  broad (FIGS. 1–4, 6) and generally dark yellow. Two wall layers are visible on the young sporogenous cell, which are continuous with OWL1 and with laminated OWL2. OWL1 is 0.4–1.0  $\mu\text{m}$  thick and semi-persistent, and OWL2 is 1.5–2.8  $\mu\text{m}$  thick and persistent as long as sc remains attached on the spore. One to (rarely) two 'hyphal pegs' are often formed on the sporogenous cells, and are 4–10  $\mu\text{m}$  thick at the sporogenous cell base tapering to 3.0–4.5  $\mu\text{m}$  within its 12–30  $\mu\text{m}$  length. Sometimes one peg continues as mycelial hypha or as a sporogenous hypha that may bear another sc in 400–800  $\mu\text{m}$  distances from the first sc.

Then, the formation of the sc can be called intercalary instead of sub-terminal. The sporogenous hypha attached to the cell is also bi-layered, 12–25 µm thick and tapering to 5–7 µm within 100–450 µm distances from the sporogenous cell. Within these distances, the sporogenous hyphal wall tapers from 1.5–2.5 µm to 1.1–1.6 µm, and 2–9 septa originating from OWL2 may be visible in the sporogenous hypha (FIG. 2).

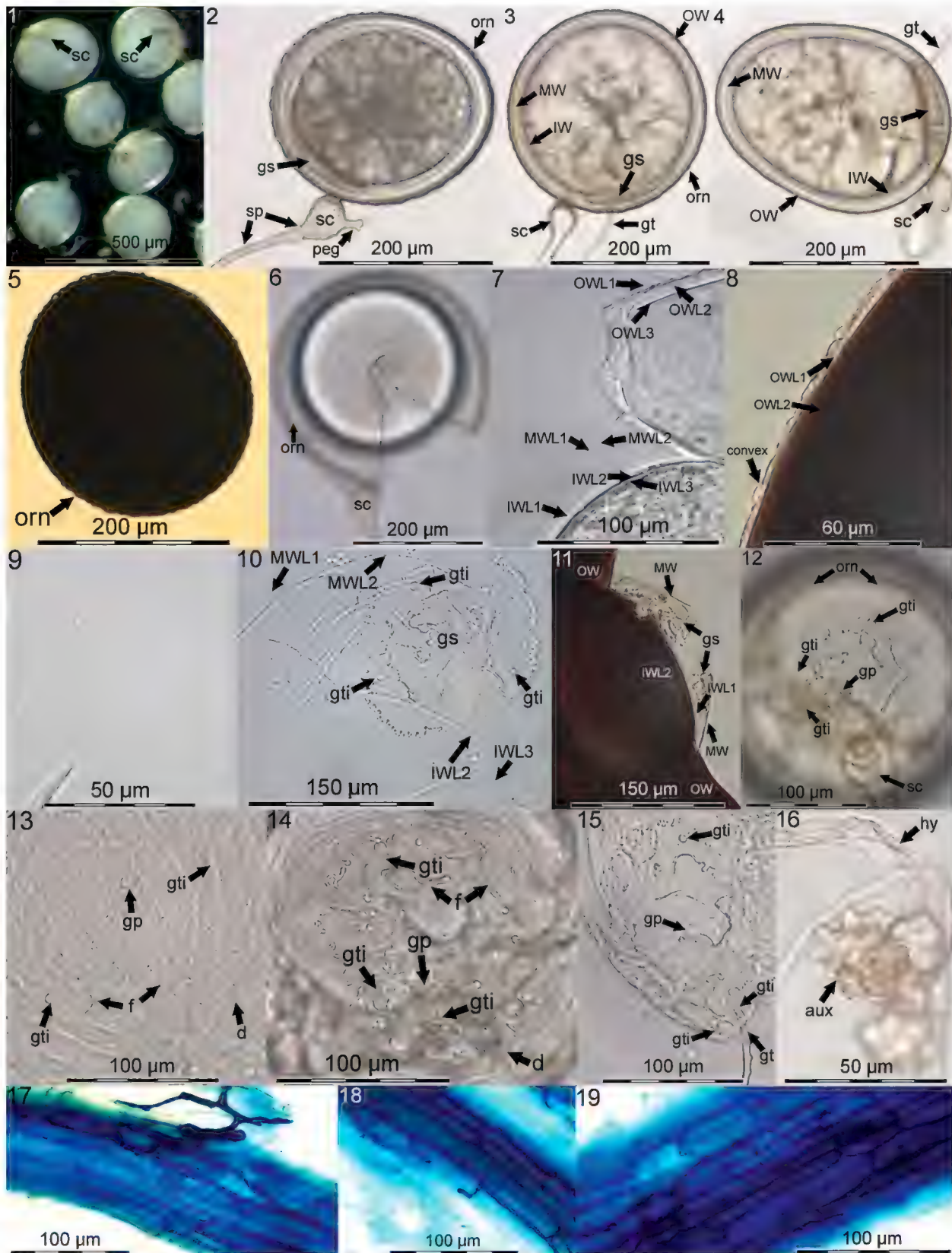
GERMINATION SHIELD is hyaline to subhyaline (FIGS. 6, 10–15), infrequently light yellow in aged spores, subglobose to oval to rarely ellipsoid, 120–125 × 135–180 µm in diameter, and generally has 6–10 lobes (FIGS. 10, 12–15). Large folds (~7–30 µm long) arise from the shield wall separating the lobes (FIGS. 10, 12–15). The one-layered shield wall and the folds are hyaline to subhyaline and generally only 0.9–1.8 µm thick. The shield periphery regularly appears slightly dentate until the germination has started. Each lobe may bear one rounded germ tube initiation (gti, FIGS. 10, 12–15), 2.5–4.5 µm in diameter. The majority of the gti's may remain undetected in young spores or in crushed mature spores due to the pressure applied on the cover slide, especially when the shields are completely separated from the spores by applying harsh pressure (FIG. 10). Single germination tubes may simultaneously emerge from 1 to 3 gti's during early germination (FIGS. 3–4, 15). They penetrate the OW (FIGS. 3–4) and branch in the spore periphery within a short distance.

SPORE DEVELOPMENT — The key stages of spore development observed in the pure and bait cultures are the same as known for other species in the *Racocetraceae*: First the outer spore wall differentiated into one semi-persistent outer layer (OWL1), the laminate, structural layer (OWL2) which differentiates the characteristic convex projections, and the adherent inner layer (OWL3). The MW and IW developed de novo with no visible connection with the outer wall. Finally, the germination shield differentiated its multiple-lobed structure, beginning from the initial germ hole (= germ pore) and forming a gti at the end

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FIGS. 1–19. *Cetraspora helvetica*. FIG. 1. White spores with pigmented sporogenous cells (sc) in a Petri dish. FIGS. 2–4. Spores have three walls: an outer, middle and inner wall (ow, mw, iw). On sc, short hyphal pegs (peg) may be differentiated, and one to several septa (sp) may be visible in the sporogenous hypha. Germination shields (gs) are formed on the outer IW surface, and sometimes germ tubes (gt) are visible in germinating spores. The convex warty projections (orn) are not obvious under low magnification in PVLG mountants. FIG. 5. Spores stain dark purple to purple black in Melzer's reagent. Here, the convex projections are conspicuous. FIG. 6. Crushed spore with focus on the ornamentation in planar view. FIG. 7. Triple-layered ow (OWL1–3), bi-layered mw (MWL1–2) and triple-layered iw (IWL1–3). FIG. 8. Laminate OWL2 stains dark purple to black purple in Melzer's reagent, while OWL1 with the convex projections (here in cross view) does not stain. FIG. 9. Ornamentation in planar view. FIG. 10. mw with thin MWL1 that slightly wrinkle, and thus, shows several folds; three germ tube initiations (gti) are in focus on the slightly crushed germ shield. FIG. 11. Crushed spore in Melzer's with gs between mw and iw; mw does not stain, while ow





and iwl2 stain dark purple to black purple. FIGS. 12–15. Germ shields (gs) in (semi-)planar view; shields have a initial germ pore (gp; = germ hole) and several lobes that are generally separated by large folds (f); the lobes may regularly bear one germ tube initiation (gti) each but the gti's often become invisible following pressure needed to present the gs in planar view or to separate the gs from overlaying ow and mw; shield periphery is slightly dentate (d) in mature spores. FIG. 16. Light yellow to yellow, knobby auxiliary cells (aux) formed on light yellow to yellow mycelial hyphae. FIGS. 17–19. Mycorrhizal structures (here roots of *Medicago truncatula*, 12 weeks after inoculation) lack intraradical vesicles.



of the shield development usually in each of the lobes; from 1–3 of these gti, the germination tubes emerge during initial germination.

**GERMINATION** — One to two germ tubes may arise. They are light yellow to bright yellow, 5–7  $\mu\text{m}$  in diameter and emerge from one or two gti's (FIGS. 3–4, 15). Germ tubes directly penetrate the ow and branch then almost immediately in the soil environment. The mono- to bi-layered germ tube walls are  $\sim 1.2\text{--}2.0$   $\mu\text{m}$  thick in close spore vicinity.

**AUXILIARY CELLS** are formed singly or in small aggregates (2–4 cells) on light yellow to yellow mycelial hyphae (FIG. 16). They are yellow, knobby and 20–25  $\mu\text{m}$  in diameter.

**ARBUSCULAR MYCORRHIZA FORMATION** is without formation of vesicles (FIGS. 17–19).

**ADDITIONAL COLLECTIONS:** **SWITZERLAND:** Kanton Bern, Langnau im Emmental, temporary grassland in April, 2009, specimens from 8 trap cultures (in July 2009; Z+ZT Myc 3040a–h); Grasswil, temporary grassland in April, 2009, specimens from 2 trap cultures (in July 2010; Z+ZT Myc 3202a–b).

**DISTRIBUTION** — *Cetraspora helvetica* has thus far been detected only at the cited locations in the Kantons Thurgau and Bern, Switzerland.

**MOLECULAR ANALYSES** — Four partial sequences of the large (LSU, 28S) subunit ( $\sim 700$  bp) of the ribosomal gene were obtained. Phylogenetic analyses firmly placed the newly described fungus into the genus *Cetraspora* adjacent to *C. spinosissima*, *C. pellucida* and *C. gilmorei* (FIG. 20). The analyses also demonstrate the monophyly of the two genera *Racocetra* and *Cetraspora* of the family *Racocetraceae* recently described (FIG. 20).

## Discussion

The three-walled glomerospores and the multiply lobed, hyaline germination shield place the newly described species in the genus *Cetraspora* in the *Racocetraceae* (Oehl et al. 2009a) of the *Diversisporales* (Schüßler et al. 2001). The molecular analyses using the 28S ribosomal gene confirmed the morphological findings: *Cetraspora helvetica* clustered in the phylogenetic tree next to *C. spinosissima*, *C. pellucida*, and *C. gilmorei*. *Cetraspora helvetica* is readily distinguished from all other known species in the *Racocetraceae* by spore color, staining features in Melzer's reagent, and the spore wall characteristics, including the characteristic convex warts on the outer spore surface.

There are only five species known within *Cetraspora* sensu Oehl et al. (2009a), i.e. species of *Scutellospora* group C sensu de Souza et al. (2005) with three spore walls and multiple-lobed germination shields. These species are: *C. armeniaca*, *C. gilmorei*, *C. pellucida*, *C. spinosissima*, and *C. striata* (Oehl et al. 2009a). However, these species have either smooth spore surfaces

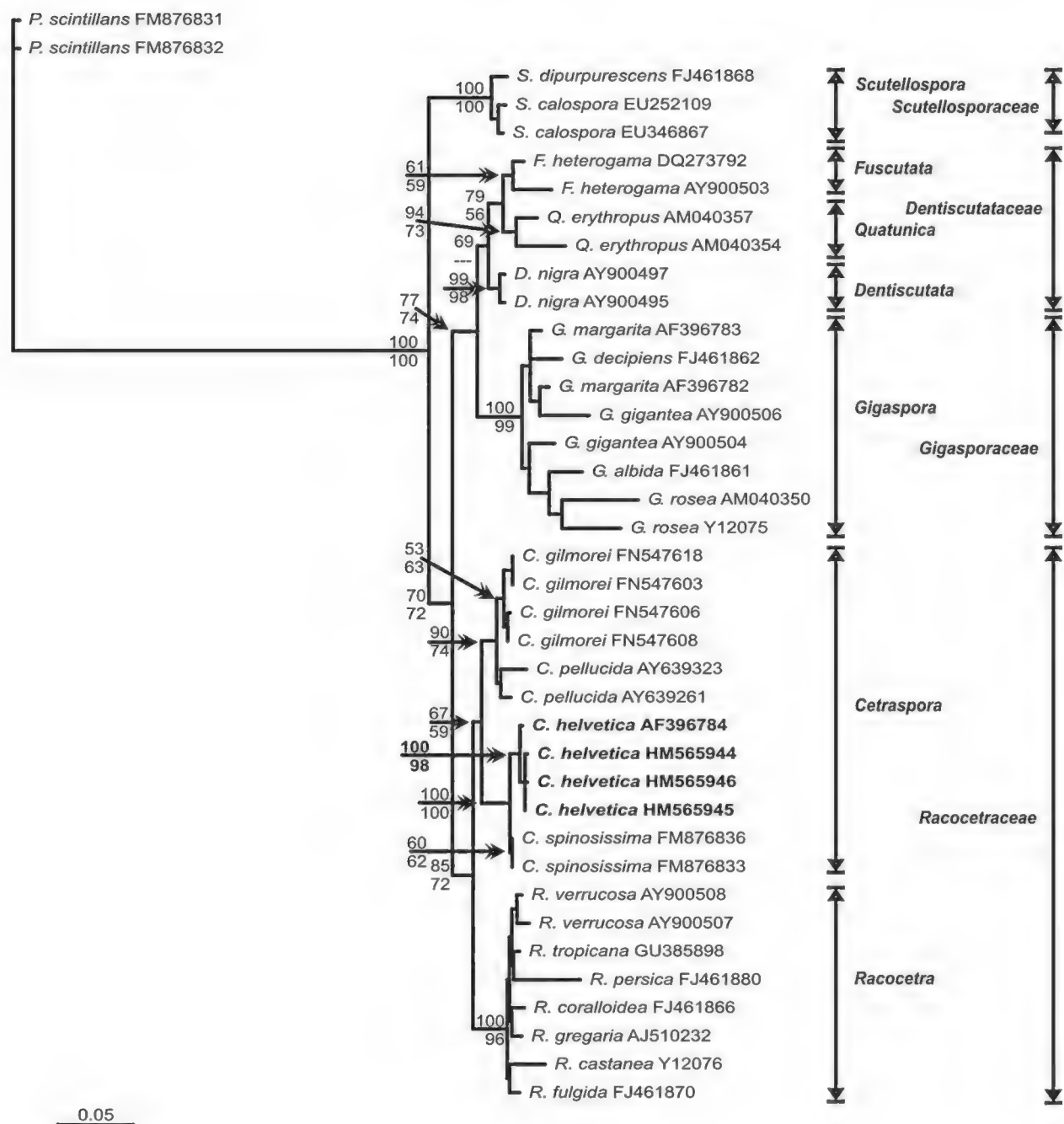


FIG. 20. Phylogenetic reconstruction of the *Gigasporineae* sensu Morton and Benny (1990) obtained from partial LSU rDNA sequences (~700 bp). The neighbor-joining (NJ) analysis was performed with GTR substitution model using the following ModelTest parameters: rate matrix (a = 0.9199, b = 10.3397, c = 2.6872, d = 0.6133, e = 21.1774); number of substitutions types = 6; nucleotide frequencies (A = 0.32210, C = 0.13470, G = 0.23930, T = 0.30390); rates = gamma; shape=0.6497 and proportion of invariable sites = 0. The four new sequences obtained are indicated in bold. Sequences are labeled with their database accession numbers. Bootstrap values (in %) are from neighbor-joining (NJ) and maximum parsimony (MP) analyses (1000 bootstraps), respectively. Only topologies with bootstrap values of at least 50% are shown. The lines to the right show the current genera and families of the *Gigasporineae*. (Consistency Index = 0.6077; Retention Index = 0.7799).

(*C. armeniaca*, *C. gilmorei*, *C. pellucida*; Błaszowski 1993, Gerdemann & Trappe 1974, Nicolson & Schenck 1979, Koske & Walker 1986) and/or do not form white spores (*C. armeniaca*, *C. spinosissima* and *C. striata*; Błaszowski 1993, Walker et al. 1998, Cuenca et al. 2008). Moreover, the ornamentations of *C. spinosissima* and *C. striata* consist of spines and fingerprint-like procedures, respectively, and not of convex warts (Walker et al. 1998, Cuenca et al. 2008).

Besides *C. helvetica*, there is only one other fungus in the *Racocetraceae* forming hyaline or white spores with a warty surface ornamentation. This is *Racocetra beninensis* Oehl et al. 2009 (Tchabi et al. 2009). However, *R. beninensis* has only two spore walls and its projections are smaller and more irregular than those of *C. helvetica*. Moreover, its inner spore wall does not stain in Melzer's reagent, while the outer wall stains bright yellow to dark yellow but not purple to dark purple as in *C. helvetica*.

Only *Scutellospora nodosa* (Błaszowski 1991), which phylogenetically belongs to *Scutellospora* group A sensu de Souza et al. (2005) and to the monogeneric family *Scutellosporaceae* sensu Oehl et al. (2009a), has a similar warty ornamentation as *C. helvetica*. However, differences in sporogenous cell color, germination shield size and structure, inner wall structure, and the staining behavior in Melzer's clearly differentiate *C. helvetica* and *S. nodosa*. *Scutellospora nodosa* has sc's that are concolorous with the spore, a simpler and substantially smaller germ shield, and an outer wall that stains brownish-red instead of dark to black purple. Additionally, of the inner wall only IWL3 stains purple in *S. nodosa*, while in *C. helvetica* it is IWL2. The IWL2 stains purple in all known *Cetraspora* spp.

It is remarkable that species of *Racocetraceae* and *Dentiscutataceae* generally have pigmented sporogenous cells (sc) even when the spore color is hyaline or white to light creamy. This is known for *R. beninensis* and *R. fulgida*, *C. pellucida*, *C. gilmorei*, and *C. helvetica*, and for *Dentiscutata cerradensis*, *D. scutata*, and *Fuscutata savannicola*, which all form light colored, hyaline to subhyaline or white spores. In *C. helvetica*, the germ tube, the mycelial hyphae, and the auxiliary cells are also concolorous with the sc, i.e. bright yellow to dark yellow. It will be interesting to determine later whether this feature is common for all (or a majority) of the *Racocetraceae* and *Dentiscutataceae* spp. Our observation is even more remarkable when considering that *Racocetraceae* spp. form hyaline to subhyaline germ shields while *Dentiscutataceae* spp. have yellow-brown to brown shields. However, the database for the mycelial hyphae and auxiliary cell morphologies is, to our knowledge, still incomplete and in need of improvement.

Notably, our study is the first to report that sporogenous cells can form not only sub-terminally, but also intercalarly. It will be interestingly to follow up in the future if this feature is unique within the *Glomeromycota*.

Our phylogenetic analyses demonstrate the monophyly of the genera *Racocetra* and *Cetraspora* in the *Racocetraceae* and fully support the analyses and classification of Oehl et al. (2009a), which have been recently criticized by Morton & Msiska (2010), who did not find major congruency between spore morphology and molecular phylogeny in this species group. In our opinion, those authors included some characters in their morphological-phylogenetic analyses that weakened their analyses. The authors also found a much higher intraspecific variability of the shields than Oehl et al. (2009a) and Oehl and co-workers who investigated the intraspecific variability of mature shields for a series of *Scutellosporaceae*, *Racocetraceae* and *Dentiscutataceae* spp (e.g. Silva et al. 2008, Tchabi et al. 2009, Goto et al. 2010, 2011, Oehl unpublished results). This discrepancy is due partly to the fact that in their attempt to include ontogeny in their analyses, Morton & Msiska considered also young, immature shields, which was not particularly helpful. Moreover, we believe that their isolates did not always derive from completely pure cultures but from oligospecies cultures — especially evident for *C. pellucida* where *Fuscutata savannicola*, *Dentiscutata scutata*, or similar species most probably co-existed in the cultures, which would invalidate the analyses and the conclusions drawn from those isolates. After investigating many specimens from several locations worldwide, we have never found brown shields in *C. pellucida*, nor have we found brown shields in the other five known *Cetraspora* spp. (e.g. Oehl et al. 2009a).

*Cetraspora helvetica* has been found thus far only in Switzerland. However, it was found in two different soil preservational agro-ecosystems — a no-till crop rotation system and two temporary grasslands that are rarely ploughed and characterized by long-interval (5–7 year) crop rotations dominated by 3–4 years of continued grass-clover production. It will be interesting to elucidate the biogeographical distribution of our new species in Switzerland and in the surrounding countries in more detail. This would be especially interesting in that the sporulation of *C. helvetica* appears to differ from that of *C. pellucida* and other sporogenous cell-forming arbuscular mycorrhizal fungi such as *S. calospora* and *G. margarita* that most commonly sporulate in late fall (e.g. Oehl et al. 2004, 2009b); in contrast, under more or less ambient light and temperature conditions, *C. helvetica* has formed spores only in early summer during our experiments (Oehl, unpublished).

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# MYCOTAXON

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## New and interesting records of lichens from Turkey

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**Abstract**—Eight species of lichenized and lichenicolous fungi are reported from the Turkish provinces of Giresun, Samsun, and Trabzon. Four taxa, *Dactylospora glaucomarioides*, *Lecania polycycla*, *Lecanora thysanophora*, and *Strigula stigmatella*, are new records for Turkey. A short description based on Turkish material is presented for each taxon.

**Key Words** —biota, biodiversity, Konakönü

## Introduction

Studies aiming to determine the lichens of the biota of Turkey have intensified in recent years (e.g. Candan & Özdemir Türk 2008, Halıcı et al. 2007, Kınalıoğlu 2009b, Öztürk & Güvenç 2010, Yazıcı et al. 2010). However large parts of the lichen biota of Turkey are still largely unknown. Until now the number of taxa recorded from different regions of Trabzon province was 518 (John 1995 [and references therein], 1999, 2000, 2002; John & Breuss 2004, John & Nimis 1998, John et al. 2000, Kınalıoğlu 2007b, 2008, Kınalıoğlu & Engin 2004, Yazıcı 1996, 1999, 2006; Yazıcı & Aslan 2002, 2005), and 431 from Giresun province (Aslan et al. 2002, Aslan & Yazıcı 2006, Duman & Yurdakulol 2007, Halıcı & Şenkardeşler 2009, John & Breuss 2004, Kınalıoğlu 2005, 2006, 2008, 2009a, Kınalıoğlu & Engin 2004, Küçük 1990, Özgen et al. 2003, Steiner 1909, Süleyman et al. 2002, Yazıcı 2006, Yazıcı & Aptroot 2008). The lichen biota of Samsun province, with 129 species reported (John et al. 2000, Kınalıoğlu 2007a, Söylemez et al. 1998), is less known than that of Trabzon and Giresun provinces. This contribution reports further species as first records for Turkey or for the provinces of Giresun, Samsun, or Trabzon.

## Materials and methods

The lichen samples were collected from the three provinces Samsun, Giresun and Trabzon between 25 August 2004 and 10 April 2010. All samples were

identified with various lichen guides (e.g. Brodo et al. 2001, Hafellner 1979, Mayrhofer 1988, Purvis et al. 1992, Wirth 1995). The specimens are deposited in the herbarium of the Faculty of Science and Arts, Giresun University, Giresun, Turkey; with some duplicates in personal herbaria of H. Sipman and T. Tønsberg. The accession numbers of the collections are given in parentheses after the locality details.

### Taxonomy

#### *Caloplaca arcis* (Poelt & Vežda) Arup

A detailed description is provided by Vondrak et al. (2009).

SPECIMEN EXAMINED: **Giresun:** Center, Ayvasıl place, sea shore, 40°55'37"N, 38°18'45"E, 10 m, 25 May 2006, on mortar, det. H. Sipman, (Kınalıoğlu 1529).

Thallus yellowish. Apothecia 0.4–0.8 mm diam., rare; thalline exciple yellow; disc orange. Ascospores ellipsoid, 10–14  $\mu\text{m}$   $\times$  5–6.5; septum 4–5 wide. Thallus K+ violet, C–, PD–.

Recently recorded as new to Turkey by Vondrak et al. (2009) from Sinop province. New to Giresun province.

Known also from Europe (Bulgaria, Italy, Netherlands, Slovakia, Spain) mainly growing on inland sun-exposed hard siliceous rocks, but also on pure limestone (Vondrak et al. 2009). In Turkey it was collected from mortar.

#### *Caloplaca limonia* Nimis & Poelt

Detailed descriptions are provided by Vondrak et al. (2009) and Nimis & Martellos (2004).

SPECIMEN EXAMINED: **Giresun:** Piraziz, Gökçeali village, 40°54'24"N, 38°05'39"E, 422 m, 30 Mar. 2006, on mortar, det. H. Sipman, (Kınalıoğlu 1608).

Thallus bright yellow. Apothecia 0.4–0.7 mm diam., numerous; thalline exciple yellow; disc orange. Ascospores ellipsoid, 12–15  $\mu\text{m}$   $\times$  5.5–8; septum 5–6.5 wide. Thallus K+ violet, C–, PD–.

Recently recorded as new to Turkey by Vondrak et al. (2009) from Çanakkale and Sinop provinces. New to Giresun province.

Known also from Bulgaria, Croatia, Czech Republic, Georgia, Italy, Morocco, Romania, Russia, Ukraine. It mainly grows on coastal calcareous rocks or base-rich, hard siliceous cliffs in dry sun exposed to shaded and damp situations, but also on twigs of maritime shrubs or on mosses and soil. It is also known from inland localities (Vondrak et al. 2009). In Turkey it was collected only from mortar.

#### *Dactylospora glaucomarioides* (Tuck.) Hafellner

A detailed description is provided by Hafellner (1979).

SPECIMEN EXAMINED: **Giresun:** Dereli, Karagöl mountains, 40°35'51"N, 38°10'30"E, 3050 m, 29 Jul. 2007, on *Ochrolechia* sp. on soil, det. H. Sipman, (Kınalıoğlu 1591).

Apothecia black, scattered on the thallus surface of the host; disc 0.1–0.5 mm diam., mostly flat, with thick margin, (0.2–0.4 mm diam.). Paraphyses septate, 1.3–2 µm thick. Ascospores dark brown, 1–3 septate, 12.5–20 × 5–7.5 µm.

New to Turkey. This lichenicolous species known also from America and Russia growing on *Ochrolechia upsaliensis* (mostly thallus, occasionally apothecia), and on *Megaspora verrucosa* (apothecia, thallus) (Hafellner 1979, Zhurbenko 2004). In Turkey, it was collected on thallus of epigeic *Ochrolechia* sp. on exposed mountain ridges.

***Lecania polycycla* (Anzi) Lettau**

Detailed descriptions are provided by CNALH (2009) and Mayrhofer (1988).

SPECIMEN EXAMINED: **Samsun**, Ayvacık, near Suatugurlu dam, 41°04'40"N, 36°40'13"E, 50 m, 22 Jul. 2006, on mortar, det. H. Sipman, (Kinalioğlu 1797).

Thallus granular or rimose to areolate, olive-brown. Apothecia abundant, 0.3–0.9 mm in diam; disc flat to slightly convex, brownish; margin whitish-gray. Hymenium 40–50 µm. Ascospores ellipsoid, 0–1 septate, 9–12.5 × 3–5 µm. Thallus C–, K–, PD–.

New to Turkey. Widespread in Europe, Africa and North America growing on calcareous rocks and rarely on acidic rocks (CNALH 2009, Mayrhofer 1988). In Turkey it was only collected from mortar in sun exposed area.

***Lecanora thysanophora* R.C. Harris**

Detailed descriptions are provided by Brodo et al. (2001), Harris et al. (2000), and Kowalewska & Kukwa (2003).

SPECIMENS EXAMINED: **Giresun**: Keşap district, Geçit village, 40°46'13"N, 38°32'48"E, 720 m, 25 Aug. 2004, on *Corylus* sp., det. Tor Tønsberg, (Kinalioğlu 1794). **Giresun**: SW of city centre, Boztekke village, 40°55'05"N, 38°18'31"E, 8 m, 10 Apr. 2010, on *Corylus* sp., det. Tor Tønsberg, (Kinalioğlu 1795).

Thallus thin, green-yellow, leprose, continuous or patchy. Apothecia not observed. Fibrous prothallus conspicuous at the thallus margins. Thallus K+ yellow, KC+ deep yellow, C–, PD–,

New to Turkey. Known also from North America and many European Countries mainly growing on trunks of deciduous trees, especially *Acer saccharum* and *Thuja occidentalis*, but also on *Populus*, *Tilia*, or even on shaded siliceous rocks, in shaded or partly shaded forest (Brodo et al. 2001, Harris et al. 2000, Kowalewska & Kukwa 2003). In Turkey it was collected from *Corylus* sp. in partly shaded and damp hazelnut gardens.

***Melanelia substygia* (Räsänen) Essl.**

Detailed descriptions (as *Melanelia tominii*) are provided by Brodo et al. (2001) and CNALH (2009).



SPECIMEN EXAMINED: **Trabzon:** Araklı, Near the Uzuntarla, 40°39'47"N 40°2'49"E, 2390, 22 Aug. 2005, on moss, det. H. Sipman, (Kınalıoğlu 1478).

Thallus dark brown to black; lobes flat to weakly convex, 1–2.5 mm wide, pseudocyphellae laminal, whitish to dark. Apothecia not observed. Medulla C+ red, KC+ red, PD–, K–.

In Turkey *Melanelia substygia* was previously recorded from Erzurum province (Yazıcı & Aslan 2000). New to Trabzon province.

Known also from Europe, North America, North Africa and Asia growing on non-calcareous rocks, usually in open, dry sites and also in forested regions (Brodo et al. 2001, CNALH 2009). In Turkey it was collected from on mosses in exposed areas at high elevation.

*Mycobilimbia berengeriana* (A. Massal.) Hafellner & V. Wirth

Detailed descriptions (as *Lecidea berengeriana*) are provided by CNALH (2009), Purvis et al. (1992), and Thomson (1997).

SPECIMEN EXAMINED: **Giresun:** Dereli, Karagöl mountains, 40°35'51" N, 38°10'30" E, 3050 m, 29 Jul. 2007, on turf, det. H. Sipman, (Kınalıoğlu 1594).

Thallus thick, white-grey, with granular warts 0.1–0.2 mm diam. Apothecia 0.3–1.2 mm diam.; disc flat or weakly convex, brownish- black. Hymenium 55–65 µm tall. Ascospores ellipsoid, 9–17 × 4–5.5 µm. Thallus C–, K–, KC–, PD–.

In Turkey previously recorded from Gümüşhane (Yazıcı & Aslan 2000) New to Giresun province.

Known also from North America, England and Scotland growing on mosses over soil and on ± calcareous rocks or on exposed turf of mountain ridges or summits (CNALH 2009, Purvis et al. 1992, Thomson 1997). In Turkey it was only collected from turf at high elevation.

*Strigula stigmatella* (Ach.) R.C. Harris

Detailed descriptions are provided by CNALH (2009), Purvis et al. (1992), and Brodo (2001).

SPECIMEN EXAMINED: **Giresun:** Dereli, Tepeköknarlı village, 40°47'28"N, 38°26'44"E, 605 m, 14. Apr. 2005, on *Carpinus* sp., conf. H. Sipman, (Kınalıoğlu 1622).

Thallus whitish-grey, very thin. Perithecia black, semi-immersed, 0.2–0.5 mm diam. Ascospores 25–36 × 5–7.5 µm, 6–7 septate, fusiform. Thallus C–, K–, KC–, PD–.

New to Turkey. Known also from the Europe, America and Canada growing on the bark of old broad-leaved trees, or over mosses on tree bases (Purvis et al. 1992). In Turkey it was only collected from on trunk of *Carpinus* sp. in entrance of shaded forest.

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## MYCOTAXON

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***Coccostromopsis palmicola* on *Butia yatay* from Argentina**

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**Abstract** — *Coccostromopsis palmicola* on living leaves of *Butia yatay* (Arecaceae) is reported for the first time from Argentina. This fungus is briefly described and illustrated. Some phytogeographical and phythopathological aspects are discussed.

**Key words** — *Ascomycota*, endangered palm, *Phyllachorales*, tar spots

**Introduction**

According to Hyde & Cannon (1999) the genus *Coccostromopsis* (*Phyllachorales*, *Phyllachoraceae*) was reintroduced for species on palms and bamboos having pulvinate, gelatinous, stromata with a yellowish sheen when young and strongly erumpent when mature, and with hyaline to yellow-brown or brown ascospores when mature. In respect to geographic distribution, species of *Coccostromopsis* are found wherever their palm hosts occur, i.e. mostly in tropical and subtropical regions (Blomberry & Rodd 1982). *Coccostromopsis* currently comprises five species. Hyde & Cannon (1999) provided a key to three of them, namely *C. diplothemii* (of which the type species, *C. palmigena*, is a synonym), *C. chamaedoreae*, and *C. palmicola*. These three species have been recorded from various countries of Central and South America. One additional species, *C. bambusae* (Sawada 1959), occurs on bamboo in China. Species of *Coccostromopsis* are considered tar spot fungi, because of the significant blackening of the surface layers of their ascomata (Hyde & Cannon 1999). The number of fungi associated with diseases of palm leaves is comparatively low, perhaps a reflection of the tough tissues of palms (Hyde & Cannon 1999). In Australia, Fröhlich (1993) identified 27 species associated with 14 palm species. Recently Capdet & Romero (2010) summarized previous information about fungi of palms and their occurrence in Argentina.

The purpose of this article is to communicate the presence of *Coccostromopsis palmicola* on living leaves of *Butia yatay* (Mart.) Becc. (*Arecaceae*) and to determine whether *C. palmicola* occurs on other palm species in Argentina.

## Materials and methods

The sampling areas comprised parts of two national parks: Iguazú in Misiones Province and El Palmar in Entre Ríos Province (Fig. 1).

Iguazú National Park covers an area of 67,620 hectares (25°41'S, 54°18'W; APN 2008). This park is included in the "Paranaense province" (Cabrera & Willink 1980) of the Argentine phytogeographical regions. The climate is subtropical without a dry season. Annual rainfall averages vary between 1600 mm and 2000 mm and the annual average temperature is 20°C. The vegetation is subtropical forest and represents the highest animal and plant biodiversity in the country (Dirección de Bosques 2003). The two palms studied in this area were *Euterpe edulis* Mart. and *Syagrus romanzoffiana* (Cham.) Glassman. El Palmar National Park, covers an area of 8,500 hectares (31°55'S, 58°14'W) and was established in 1965 with the aim of preserving *Butia yatay*, an endangered species (Chebez 1994). It is included in the Argentine phytogeographical region called "Espinal province" (Cabrera & Willink 1980). The climate is warm and humid in the north, and temperate and dry in the west and south. Rainfall ranges from 400 mm to 1500 mm, mainly in spring and summer (Dirección de Bosques 2003). The vegetation includes savanna with palms, shrubs and gallery forest along the Uruguay River and grasslands. *Butia yatay*, the only palm present in the Park, has an endemic distribution in southern South America occurring in Argentina, Brazil, Paraguay and Uruguay.

Intensive collecting was conducted in El Palmar National Park over the past three years (2007-2009). Living leaves of palm were collected in different seasons. The material was air-dried. Microscopic characters were observed in vivo using light microscopy. Sizes of all the structures were based on 20 measurements. Drawings were made with a camera lucida. Photographs were taken with a Sony Digital camera. The specimens are deposited in the BAFC fungal reference collection (Holmgren et al. 1990).

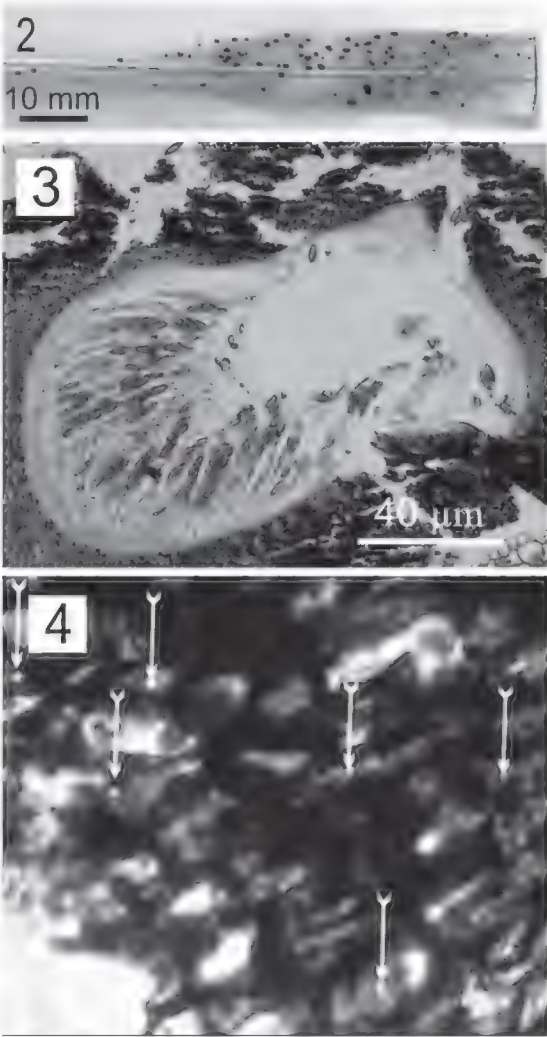
## Results

No specimens were found on *Euterpe edulis* or *Syagrus romanzoffiana* in Iguazú National Park. In contrast, most of the leaf pinnae of the palms trees observed of *Butia yatay* in the El Palmar National Park, Entre Ríos, had many stromata along the length of the leaflet.

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FIGS 1–8. 1. Sampling sites. 2. Stromata on pinna of *Butia yatay*; scale bar = 10 mm. 3. Longitudinal section through a perithecial ascoma; scale bar = 40 µm. 4. Peridium cells with Munk pores; scale bar = 10 µm. 5. Young stroma on host surface; scale bar = 0.25 mm. 6. Teleomorphic stroma on leaf; arrow points to black ascospore mass; scale bar = 0.25 mm. 7. Stroma with conidioma; arrow points to caramel brown conidial mass with cerebriform aspect; scale bar = 0.5 mm. 8. Stroma with hyperparasitic conidioma; arrow points to translucent white cirrus; scale bar = 1 mm.





*Coccostromopsis palmicola* (Speg.) K.D. Hyde & P.F. Cannon,

Mycol. Pap. 175: 67, 1999.

FIGS. 2–14

= *Auerswaldia palmicola* Speg., Anal. Soc. cient. argent. 19: 247, 1885. Type LPS 277!

ADDITIONAL SYNONYMY: see Hyde &amp; Cannon (1999).

STROMATA 1.8–2.7 mm long, 1–1.6 mm wide, on living leaves, distributed along the veins, primarily on adaxial surface but also present on abaxial surface, with a sulphur-yellow patina when young, usually hemispherical or elongated, erumpent, verrucose, opaque black with shiny black areas formed by ascospore mass when mature. Cells of the stroma with Munk pores.

TELEOMORPH. ASCI cylindric-clavate, apex truncate, 8-spored,  $120\text{--}155 \times 16\text{--}25\ \mu\text{m}$ , long-stalked,  $35\text{--}45\ \mu\text{m}$  long. ASCOSPORES  $25\text{--}28 \times 8\text{--}11\ \mu\text{m}$ , arranged multiseriately, guttulate, aseptate, fusiform-ellipsoidal, mid brown, surrounded by a mucilaginous sheath. ANAMORPH. CONIDIOMATA formed locules in upper part of stroma, irregularly shaped. CONIDIOGENOUS CELLS in cluster on short branched conidiophores, cylindrical, enteroblastic. CONIDIA  $14\text{--}31 \times 1\text{--}2\ \mu\text{m}$ , filiform, round towards both ends, often curved, aseptate, smooth, hyalines. Some of the stromata are parasitized by an anamorph producing conidiomata inside the stroma with a white cirrus consisting of fusiform to flabelliform conidia,  $9\text{--}14 \times 2\text{--}3\ \mu\text{m}$ .

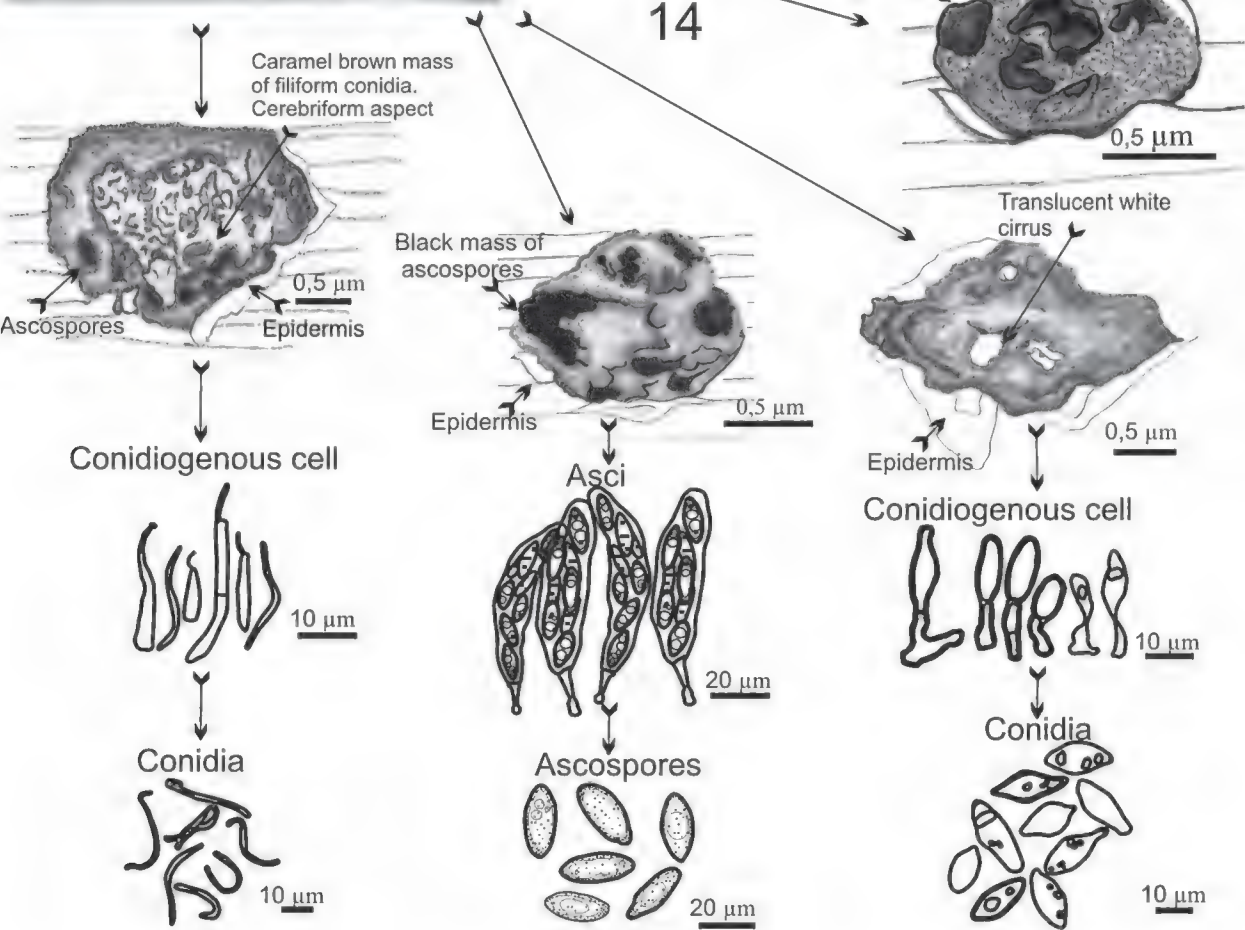
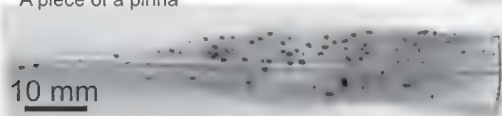
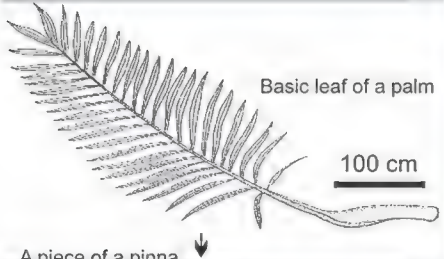
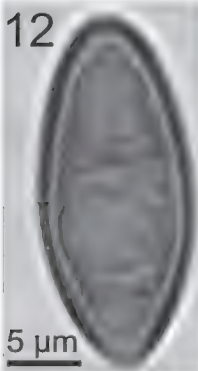
MATERIAL EXAMINED — ARGENTINA. ENTRE RÍOS, DPTO. COLÓN: EL PALMAR NATIONAL PARK, coll. Cabral, D., Iannone, L. & Pereira, S. 22.II.2007 (BAFC 51782), 22.II.2007 (BAFC 51783); 23.II.2007 (BAFC 51784); 24.II.2007 (BAFC 51780); coll. Capdet, M. & Romero, A.I. 22.IV.2008 (BAFC 51779); 23.IV.2008 (BAFC 51778); 24.IV.2008 (BAFC 51777); 18.VIII.2008 (BAFC 51785); 20.VIII.2008 (BAFC 51785); 02.II.2009 (BAFC 51781).

NOTES: This is the first record of *Coccostromopsis palmicola* on *Butia yatay* from Argentina. Spegazzini (1885) originally described this fungus on leaves of *Butia yatay* from Paraguay. Later Viégas (1944) reported it from Brazil on leaves of *Allagoptera arenaria* (Gomes) Kuntze. Although the collections from Paraguay and Brazil were collected in springtime, we have found it during all the seasons, although the summer collections were in the best condition. Of the 50 *Butia yatay* trees observed in different parts of the park, all were infected with *C. palmicola*.

Knowing that the fungus occurs in Brazil on other palm species, we also looked in Iguazú National Park close to the boundary with Brazil. *Butia yatay* is not present in Misiones province (Cabral & Castro 2007), but we examined two palms: *Euterpe edulis* and *Syagrus romanzoffiana* that grow in Brazil and Paraguay (Cabral & Castro 2007). *Coccostromopsis palmicola* was not found on these hosts. How can we explain its presence in Paraguay and Argentina

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FIGS 9–14. 9–11. Asci; arrow indicates apex details; scale bars =  $10\ \mu\text{m}$ . 12–13. Ascospores; scale bars =  $5\ \mu\text{m}$ . 14. General outlines of the different morphologies found on pinnae of *Butia yatay*.





in El Palmar National Park? As observed in FIG. 1, in Argentina there are two main riverine systems: the Uruguay riverine system and the Paraguay-Paraná riverine system, which is a 3400 km long natural corridor through various ecosystems (tropical rain forest, savannas, steppes and brushlands) between 16 and 34° of south latitude (Neiff et al. 2005). The Uruguay system connects Brazil-Misiones to Entre Ríos provinces. The National Park of Entre Ríos is on the Uruguay side of the river side east of the province while the Paraná River is on the west side of the province. Therefore, the fact that Paraguay is connected through the Paraná River with the Entre Ríos province explains the presence of *C. palmicola* in both sites.

We cannot answer the question why *C. palmicola* is not in Misiones province, which shares the climate and most of the flora with Brazil and Paraguay. Although we did not find *C. palmicola* on *Euterpe edulis* and *Syagrus romanzoffiana*, we cannot say that the fungus is host specific on *Butia yatay* because in Brazil it is found on *Allagoptera arenaria* (Viégas 1944).

In our results we mentioned above that the stromata were parasitized by another anamorph. In his revision of *Phyllachoraceae*, Cannon (1991) noted that members of this family are among the most heavily parasitized fungi.

### Acknowledgments

This study was supported by Consejo Nacional de Investigaciones Científicas y Técnicas (Conicet) (PRHIDEB Publication N° 184) and by the UK Darwin Initiative. We thank Aristóbulo Maranta from El Palmar National Park (Entre Ríos) and his team, and also to technician Gabriela Zarlavsky for the histological microscopic sections. We want to thank the expert reviewers, Dr. Amy Rossman and Dr. Paul Cannon, for their efforts to improve this paper.

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## MYCOTAXON

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**Morphological studies of *Hyphoderma cremeoalbum*  
and *Radulomyces roseolus***

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**Abstract** — Type studies reveal that *Radulomyces roseolus* is conspecific with *Hyphoderma cremeoalbum* (Basidiomycota, Polyporales). Embedded, fusoid cystidia and haplohyphidia are critical diagnostic features of *H. cremeoalbum*. Known from Europe, United States, Argentina, and New Zealand, its preferred substrate is decorticated and decayed gymnospermous wood, especially *Picea*, but the species also occurs on woody angiosperms.

**Key words** — Corticiaceae sensu lato, *Corticium cremeoalbum*, phlebioid clade, taxonomy

**Introduction**

*Hyphoderma* Wallr. is a genus of ubiquitous corticioid homobasidiomycetes with about 100 species reported worldwide (Parmasto et al. 2004). An old but vaguely circumscribed genus, recent molecular studies demonstrate that *Hyphoderma* is polyphyletic with most species distributed in two clades — the *Hymenochaetales* and the *Polyporales* (Langer 2001; Larsson 2007; Larsson et al. 2004). Larsson (2007) resurrected the genus *Peniophorella* P. Karst. to accommodate most of the *Hyphoderma* species in the *Hymenochaetales*. *Hyphoderma* sensu stricto, in the *Polyporales*, consists of species with resupinate, effuse basidiomes, monomitic hyphal systems of clamped hyphae, often with leptocystidia or other types of cystidia, suburniform to subcylindrical basidia with four sterigmata, and thin-walled, smooth basidiospores that range from cylindrical to subglobose (Larsson 2007).

*Radulomyces roseolus* (Parmasto 1968), known only from the type from Georgia in eastern Europe, is morphologically similar to *Hyphoderma cremeoalbum*. In this study, type specimens of *Corticium cremeoalbum* and *R. roseolus* were examined and determined to be conspecific. The types are

described, illustrated, and compared, and a description of *H. cremeoalbum* is provided.

### Materials and methods

Thin, freehand sections or scrapings from the basidiomes were mounted in a Melzer's reagent (Kirk et al. 2008) or 1% (weight/volume) aqueous phloxine and 1% (w/v) aqueous potassium hydroxide. Drawings were made with a camera lucida attachment on an Olympus BH2 compound microscope. Q values were obtained from dividing average basidiospore length by width (Kirk et al. 2008). Basidiospores are often scarce in specimens, thus Q values based on less than 30 basidiospores are approximate and indicated with an asterisk (\*). Color names are from Kornerup & Wanscher (1978), and herbarium designations follow that of Index Herbariorum (Thiers, continuously updated).

The term "haplohyphidia" refers to the simple, unbranched, unmodified hyphal ends developed in the hymenium (Donk 1964). Although little used, this term is useful to distinguish among the various types of hyphidia produced in corticioid fungi.

### Taxonomy

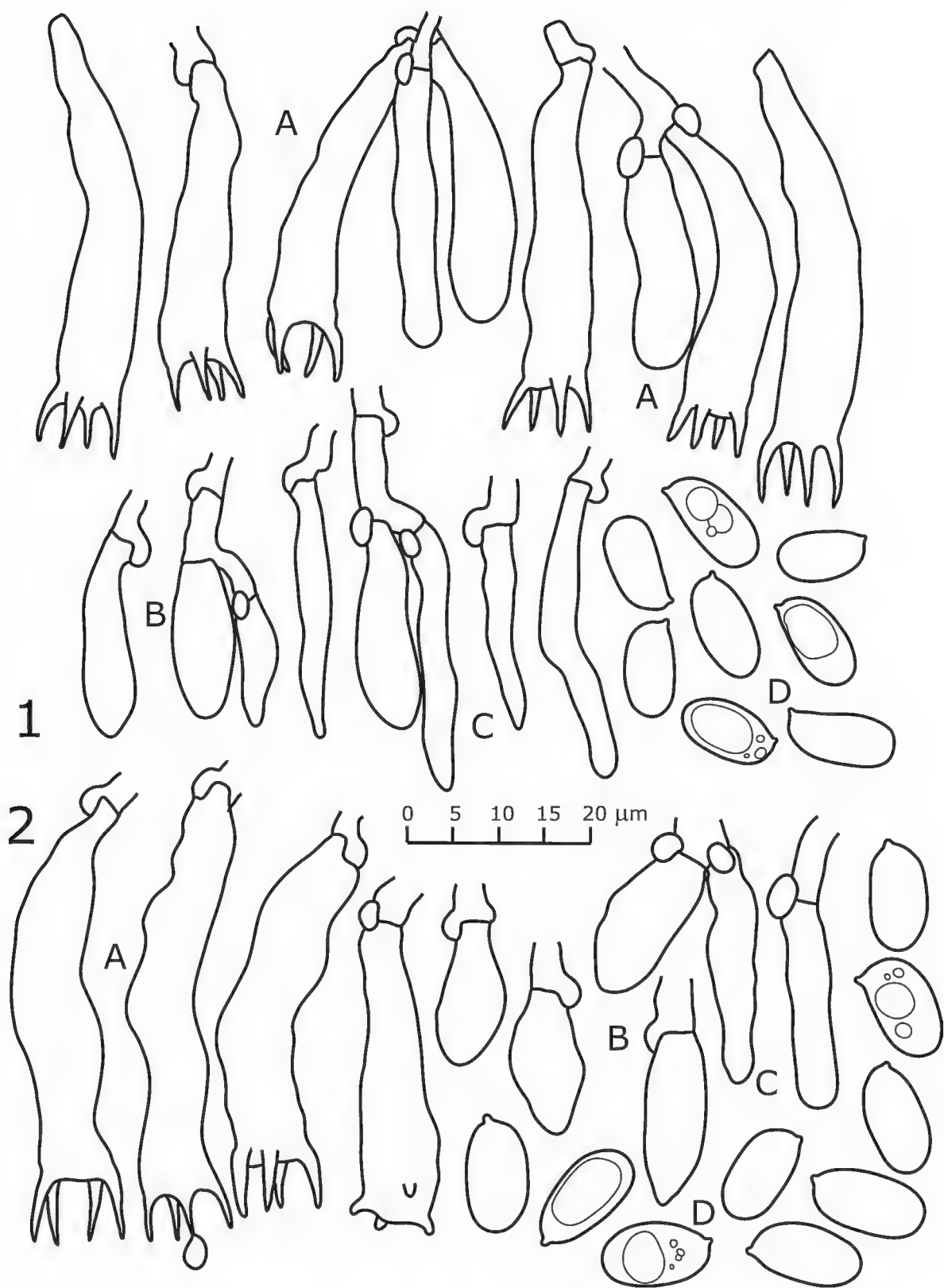
#### Type species descriptions

*Radulomyces roseolus* Parmasto, Consp. syst. cortic. p. 222. 1968.

FIG. 1

HOLOTYPE: RPSS. Georgica: Hulo in piceeto, ALT. 1300 M., ad caudicem Piceae orientalis prolapsum, 7 October 1963, E. Parmasto (TAA 16822).

BASIDIOME resupinate, effuse, colonies irregular, up to 8 × 6 mm, thin, up to 100 µm thick, subceraceous to submembranous. HYMENIAL SURFACE smooth, pruinose, yellowish white (4A2), orange white [5A(2–3)], or greyish orange (5B3). MARGIN thinning out, pruinose, concolorous with hymenial surface or white to off-white. HYPHAL SYSTEM monomitric with clamped generative hyphae. SUBICULUM indistinct, up to 30 µm thick; subicular hyphae 3.5–5.5 µm diam, clamped, moderately branched, walls thin, hyaline, smooth. SUBHYMENIUM up to 30 µm thick, a dense, compact tissue; subhymenial hyphae similar to subicular hyphae. HYMENIUM up to 50 µm thick, a dense palisade of haplohyphidia, cystidia, and basidia. HAPLOHYPHIDIA embedded, numerous, cylindrical, tapering slightly toward apex, (13–)23–40 × 3–4 µm, clamped at base, simple, unbranched, walls thin, hyaline, smooth. CYSTIDIA embedded, inconspicuous, clavate to broadly fusiform with an obtuse apex, 17–22 × 5.5–7.5 µm, clamped at base, walls thin, hyaline, smooth. BASIDIA clavate, 28–45(–55) × 6.5–8(–9) µm, clamped at base, walls thin, hyaline, smooth; 4-sterigmate. BASIDIOSPORES cylindrical, (9.3–)10–12(–13) × 5–6 µm, average of 16 spores 10.9 ± 1.0 × 5.6 ± 0.3 µm, Q = 1.9\*, with oil-like globules, walls thin, hyaline, smooth, acyanophilous, not reacting in Melzer's reagent.



FIGS. 1–2. Line drawings of microscopic elements.  
1. *Radulomyces roseolus* holotype (TAA16822). 2. *Corticium cremeoalbum* holotype (Höhnelt 684).  
A, basidia; B, cystidia; C, haplohyphidia; D, basidiospores.

COMMENTS — In the type, the well-decayed wood is broken up into fragments that support even smaller fragments of the basidiome. On one fragment is a brown-colored basidiome, which represents a *Tomentella* species, probably *T. sublilacina* (Ellis & Holw.) Wakef. Observations of the type correlate closely to the protologue except for minor differences. For example, the basidiospores observed were slightly smaller than originally cited  $10\text{--}14(-15) \times 5.5\text{--}6.5$  ( $-7$ )  $\mu\text{m}$  — and yellow resinous materials in the hymenium and subiculum described in the protologue were not observed. In addition, the hymenium color is described as “incarnato-roseum, nonnumquam cremeo coloratum”, but pink-colored hymenia were not observed in the type material, possibly because the pink color of fresh specimens fades to cream in dried material. Although the presence of haplohyphidia probably led to the placement of this taxon in *Radulomyces*, most *Radulomyces* species have thicker, robust basidiomes with distinct tubercles or spines. In a note included in the type envelope, B. Duhem noted a similarity of *R. roseolus* with *H. cremeoalbum* and suggested that they were conspecific.

***Corticium cremeoalbum*** Höhn. & Litsch., Wiesner-Festschrift p. 63. 1908. FIG. 2

HOLOTYPE: (AUSTRIA) Wiener Wald, am Sattelberg bei Preßbaum, auf morschem Nadelholz, 2 October 1901, Höhnelt no. 684 (FH 00258439).

BASIDIOME resupinate, widely effuse, thin, up to 75  $\mu\text{m}$  thick, subceraceous to membranous. HYMENIAL SURFACE discontinuous, smooth to slightly uneven with barely differentiated warts, pruinose to porulose, yellowish white (4A2) to greyish yellow (4B3). MARGIN indistinct, thinning out, pruinose, concolorous with hymenial surface. HYPHAL SYSTEM monomitic with clamped generative hyphae. SUBICULUM up to 40  $\mu\text{m}$  thick, composed of partially agglutinated hyphae arranged perpendicular to substrate; subicular hyphae 5–7  $\mu\text{m}$  diam, clamped, moderately branched, walls thin, hyaline, smooth. SUBHYMENIUM indistinct. HYMENIUM up to 40  $\mu\text{m}$  thick, a dense palisade of haplohyphidia, cystidia, and basidia. HAPLOHYPHIDIA embedded, scattered, cylindrical or tapering slightly toward apex,  $23\text{--}25 \times 5$   $\mu\text{m}$ , clamped at base, simple, unbranched, walls thin, hyaline, smooth. CYSTIDIA embedded, scattered, broadly fusoid to ovoid,  $16\text{--}21 \times 6\text{--}8.5$   $\mu\text{m}$ , clamped at base, walls thin, hyaline, smooth. BASIDIA more or less cylindrical with slight, irregular constrictions or clavate,  $30\text{--}55 \times (6.5\text{--})8\text{--}10$   $\mu\text{m}$ , clamped at base, walls thin, hyaline, smooth; 4-sterigmate. BASIDIOSPORES broadly cylindrical  $(9.5\text{--})10\text{--}12(-13) \times 6\text{--}7$   $\mu\text{m}$ , average of 20 spores  $11.5 \pm 0.8 \times 6.3 \pm 0.3$   $\mu\text{m}$ ,  $Q = 1.8^*$ , with oil-like globules, walls thin, hyaline, smooth, acyanophilous, not reacting in Melzer's reagent.

COMMENTS — The type of *C. cremeoalbum* is in good condition. The protologue, however, does not mention the presence of haplohyphidia or fusoid cystidia. Basidiospore length, given in the protologue as  $10\text{--}14 \times 5.5\text{--}6.5$   $\mu\text{m}$ ,



is slightly longer than observed. Except for these differences, the type does not deviate significantly from the protologue. Litschauer's specimen, mislabeled as holotype in Eriksson & Ryvarden (1975, p. 464), differs from the holotype at FH in lacking cystidia. Haplohyphidia are illustrated but interpreted as immature basidia.

No significant discrepancies were observed between the types of *R. roseolus* and *C. cremeoalbum*. In fact, the morphological similarities are overwhelming, and one can only conclude that these taxa are conspecific. An additional 25 herbarium specimens of *H. cremeoalbum* were studied to provide the expanded and inclusive description below.

### Species description

*Hyphoderma cremeoalbum* (Höhn. & Litsch.) Jülich, Persoonia 8(1): 80. 1974.

FIG. 3

= *Corticium cremeoalbum* Höhn. & Litsch., Wiesner-Festschrift p. 63. 1908.

= *Radulomyces roseolus* Parmasto, Consp. syst. cortic. p. 222. 1968.

= *Cerocorticium roseolum* (Parmasto) Jülich & Stalpers, Verh. Kon. Ned. Akad. Wetensch., Afd. Natuurk. II 74: 72. 1980.

BASIDIOME resupinate, widely effuse, thin, up to 200 µm thick, subceraceous to membranous. HYMENIAL SURFACE smooth to slightly uneven, sometimes verruculose, up to 3 warts per mm, sometimes discontinuous, porulose to pruinose or subfelty, yellowish white [(2–4)A2], dull yellow [3B3], pale yellow [4A2], orange white [5A(2–3)], yellowish grey [4B2], greyish yellow [(4–5)B3], pale orange [5A3], or greyish orange [(5–6)B3], warts occasionally discolored brown. MARGIN thinning out, indistinct, pruinose. HYPHAL SYSTEM monomitic with nodose-septate generative hyphae. SUBICULUM up to 150 µm thick, a moderately dense tissue of partially agglutinated ascending hyphae and coarse, hyaline crystal clusters; subicular hyphae 3.5–7 µm diam, occasionally inflated up to 11 µm diam at nodes, clamped, moderately to frequently branched, walls thin, hyaline, smooth. SUBHYMENIUM indistinct, up to 30 µm thick, a moderately dense tissue of partially agglutinated, short-celled hyphae; subhymenial hyphae 4–8 µm diam, clamped, frequently branched, walls thin, hyaline, smooth. HYMENIUM up to 50 µm thick, a dense palisade of haplohyphidia, cystidia and basidia. HAPLOHYPHIDIA scattered to numerous, cylindrical or tapering slightly toward apex, (16–)22–35(–48) × 3–6 µm, clamped at base, simple, rarely branched, walls thin, hyaline, smooth. CYSTIDIA enclosed, scattered, broadly fusoid to ovoid, rarely globose, 14–28 × 6–14 µm, clamped at base, walls thin, hyaline, smooth. BASIDIA clavate, suburniform to subcylindrical with slight, irregular constrictions, (23–)30–55 × 6.5–10.5 µm, clamped at base, walls thin, hyaline, smooth; 4-sterigmate. BASIDIOSPORES broadly cylindrical to cylindrical, (9.5–)10–14(–17) × 5–7(–8) µm, average size 11.6–13.4 × 5.5–6.6

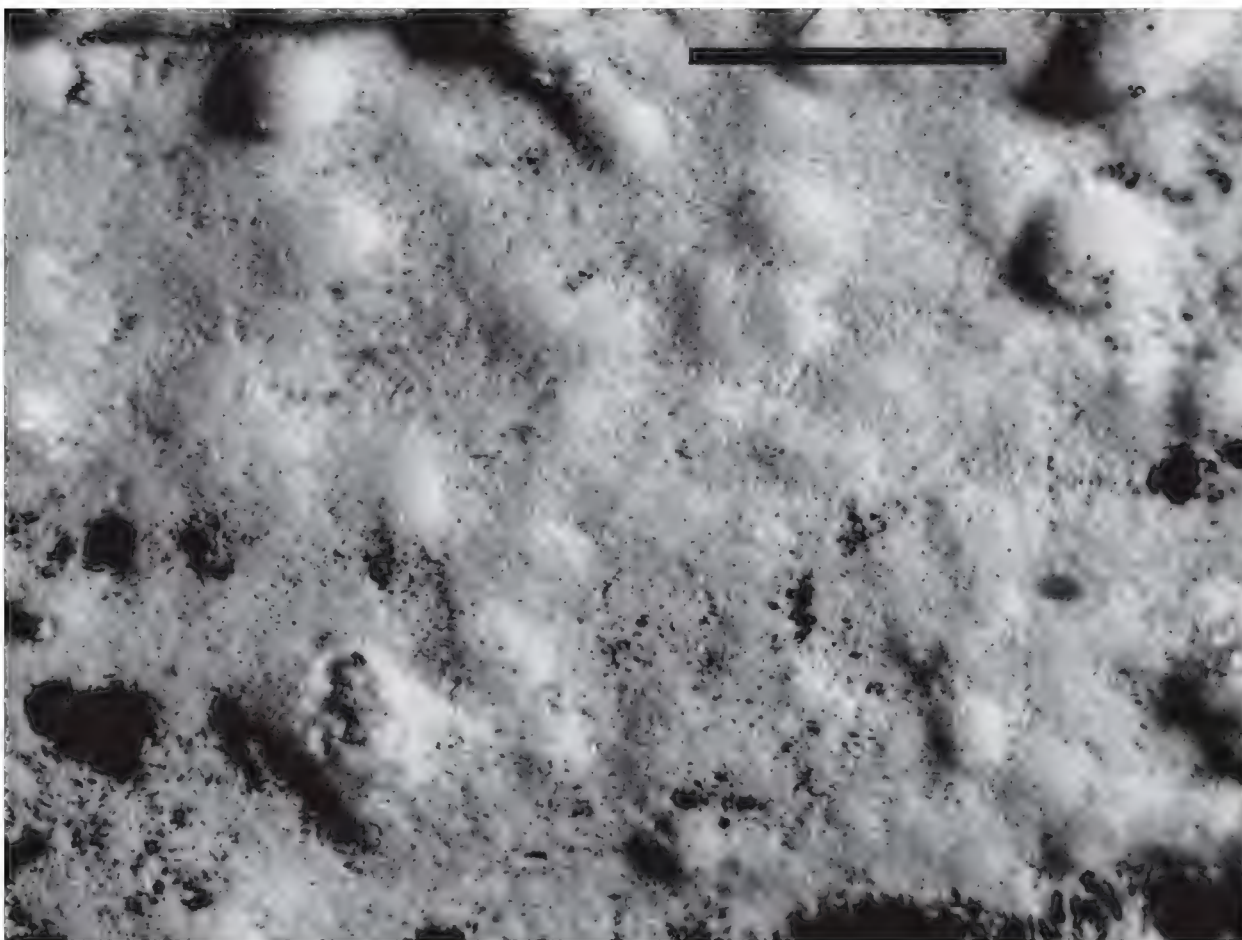


FIG. 3. Verruculose basidiome surface of *Hyphoderma cremeoalbum* (KHL4100). Bar = 1 mm.

$\mu\text{m}$ ,  $Q = 2^*-2.1$ , often containing oil-like globules, occasionally germinating, walls thin, hyaline, smooth, acyanophilous, not reacting in Melzer's reagent.

**HABITAT** — Well-decayed wood and bark of gymnosperms, especially *Picea*, and angiosperms.

**DISTRIBUTION** — Argentina, Austria, Finland (Kotiranta & Larsson 1989), France, Georgia, Germany (Grosse-Brauckmann 1990), Italy, New Zealand, Norway, Romania, Spain (Hjortstam et al. 1981, Tellería 1990), Sweden, Switzerland, Turkey, United States (Washington).

**HOLOTYPE SPECIMENS EXAMINED** — *Radulomyces roseolus* and *Corticium cremeoalbum* – see above.

**REPRESENTATIVE SPECIMENS EXAMINED** — **ARGENTINA.** DEPARTAMENTO CHUBUT: Languiño, Lago Guacho, on (well-decayed) *Nothofagus pumilio* (Poepp. & Endl.) Krasser, 18 April 1997, A Greslebin 807; Tierra del Fuego, DEPARTAMENTO USHUAIA: Estancia El Valdéz, on (well-decayed) *N. pumilio*, 04–05 March 1996, A. Greslebin 225 and 354. **AUSTRIA.** Salzburg, Hohe Tauern, Taxenbach, 1200 m s.m., on *Picea abies* (L.) H. Karst., 13 July 1997, W. Dämon, RP96/257E (Herb. Dämon); Kalkalpen, Golling, 1500–1600 m s.m., on *P. abies*, 10 August 1997, W. Dämon, RP96/257G (Herb. Dämon). **FRANCE.** Forêt de Fontainebleau, Gorge aux Loups, parcella 527, on decayed trunk of *Fagus sylvatica* L., 31 October 2006, E. Martini 9490 (Herb. Martini). **Italy.** Riserva di

Sesso Fratino (FC), 720 m, on *Abies alba* Mill., 10 October 1991, A. Bernicchia 5651 (HUBO). **NEW ZEALAND.** Bay of Plenty, Te Waiiti, on decaying wood (bark), 17 May 2006, B.C. Paulus and P.R. Johnston, BCP3640, PDD89111 (PDD). **NORWAY.** Hedmark, Lötén, Gitvola, on well-decayed, decorticated *Picea* log, 11 September 1986, K.-H. Larsson 6508, GB1773, GB0052597 (GB). **ROMANIA.** Neamt, Monastery Sihastria, in *Fagus* forest, on decayed, decorticate *Fagus* log, 17 October 1985, N. Hallenberg 9216, GB1549, GB0052600 (GB). **SWEDEN.** DALARNA: Särna Parish, Fulufället at Gjöjan, close to Falun, on (decorticate, decayed) *Picea abies* 10 September 2004, K.-H. Larsson 12404, GB0052601 (GB); LYCKSELE LAPPMARK: Sorsele Parish, Grannäs, Västra Lairobäcken, on timber at abandoned saw mill, 28 August 1983, K.-H. Larsson 4110, GB885, GB0052598 (GB); Lycksele Lappmark Kirjesålandet, Vittertj. in alpine *Picea-Betula* forest, on stem of *Betula*, 16 August 1982, K.-H. Larsson 2677, GB456, GB0052766 (GB). **SWITZERLAND.** (TESSIN) Malvaglia, on decayed coniferous wood, 19 September 1987, E. Martini 1206 (herb. Martini); (TESSIN) Meride, Bagno, on decayed, decorticated *Tilia cordata* Mill., 2 June 2007, E. Martini 9834 (Herb. Martini). **TURKEY.** NE Anatolia, Trabzon area, Sumela Monastery, on (decorticate, decayed) *Picea* wood, 2 October 1989, N. Hallenberg 11538, GB2270, GB0052599 (GB). **UNITED STATES.** WASHINGTON: Olympic National Forest, Quinault Research National Area, Plot 10-1-A-5, on decayed *Picea sitchensis* (Bong.) Carrière log, 15 October 1992, H.H. Burdsall, Jr. and M. Banik, HHB14826 (CFMR); Plot 10-1-A-13, on bark of *P. sitchensis*, 15 October 1992, H.H. Burdsall, Jr., HHB14834 (CFMR).

COMMENTS — *Hyphoderma cremeoalbum* is characterized by thin, smooth to verruculose basidiomes, cylindrical basidiospores, haplohyphidia, and enclosed fusoid cystidia. Because the cystidia are enclosed in the hymenium and haplocystidia are barely differentiated, they are easily overlooked. The description and illustrations of *H. cremeoalbum* in Eriksson & Ryvarden (1975) does not include information on cystidia, and haplohyphidia are interpreted as developing basidia. Hallenberg (1991) found that haploid isolates of *H. cremeoalbum* from Norway, Sweden, Turkey and Romania were partially or fully compatible. Although most frequently collected in Europe, *H. cremeoalbum* is widely distributed as evidence by collections from northwestern United States, southern Argentina (Greslebin 2002, Greslebin & Rajchenberg 2003), and New Zealand.

There are three species of *Hyphoderma* morphologically similar to *H. cremeoalbum*. In *Hyphoderma nemorale* K.H. Larss. and *H. incrustatum* K.H. Larss., the cylindrical basidiospores are slightly narrower ( $Q = 2.55$  and  $2.57$ , respectively) than in *H. cremeoalbum*. Additionally, they produce large, cylindrical, embedded cystidia as well as capitate or subcapitate hymenial cystidia (Larsson 1998). Like *H. cremeoalbum*, *H. sibiricum* (Parmasto) J. Erikss. & Å. Strid has haplohyphidia but significantly smaller basidia,  $25\text{--}35\text{--}(40) \times 5\text{--}7 \mu\text{m}$ , and basidiospores,  $7\text{--}8\text{--}(9) \times (4\text{--})4.5\text{--}5 \mu\text{m}$  (Eriksson & Ryvarden 1975; Ginns 1982).

*Hyphoderma cremeoalbum* was reported on *Quercus ilex* L. from Sardinia, AB6632 (Bernicchia et al. 2008); however, this specimen appears to be



*H. malenconii* (Manjón & G. Moreno) Manjón et al. Jung (1987) cited two specimens of *H. cremeoalbum* from southeastern United States on *Abies fraseri* (Pursh) Poir. but neither is correctly identified. TENN 46846 is probably *H. pilisetum* (Burt) Liberta. In TENN 46975, the basidiospores are narrower than typical for *H. cremeoalbum*; this specimen appears to represent *H. occidentale* (D.P. Rogers) Boidin & Gilles. From Arizona, Gilbertson & Bigelow (1998) reported *H. cremeoalbum*, RLG 16887, on *Pseudotsuga menziesii* (Mirb.) Franco, but this specimen is *Peniophorella praetermissa* (P. Karst.) K.H. Larss. Gilbertson et al. (2002) listed *H. cremeoalbum* from Moloka'i, Hawaii, on *Eucalyptus robusta* Sm. The specimen, RLG 22966, has numerous fusoid gloeocystidia and appears to be an undescribed species with close affiliation to *P. praetermissa*. The report of *H. cremeoalbum* from the Leningrad region on *Populus tremula* L. should be reconfirmed because cystidia and haplohyphidia were not observed (Zmitrovich & Spirin 2002). Similarly, reports of *H. cremeoalbum* from Italy on *Castanea sativa* Mill. (Mayrhofer et al. 2001) and from China (Maekawa & Zang 1995, Maekawa et al. 2002), need to be confirmed.

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The curators of the following herbaria arranged specimen loans: ARIZ, BPI, FH, GB, HUBO, PDD, TAA, TENN. Elia Martini of Bignasco, Switzerland, and Dr. Wolfgang Dämon of Salzburg, Austria, loaned specimens from their private herbaria. Drs. Harold H. Burdsall, Jr. and Wolfgang Dämon reviewed an earlier draft of this manuscript and provided valuable comments and corrections.

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# MYCOTAXON

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## Taxonomic studies of *Alternaria* from Russia: new species on *Asteraceae*

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**Abstract** — Two new species are added to the 32 *Alternaria* species known on plants of *Asteraceae*. The newly described species are *A. silybi* from *Silybum marianum* and *Alternaria simmonsii* from *Sonchus* sp.

**Key words** — milk thistle, sow thistle

### Introduction

There are 32 accepted *Alternaria* species known on plants of *Asteraceae* (Simmons, 2007). Most of them (24) belong to the group of large-spored species characterized by relatively long conidia with filiform beak. Usually they are pathogenic and have strong host specialization. During the study of mycobiota of weeds and wild herbaceous plants we have obtained a few isolates of two new *Alternaria* species on leaves of milk thistle and sow thistle. The leaves collected had a number of spots and abundant sporulation of *Cercospora* sp. on leaves of milk thistle and *Septoria sonchifolia* Cooke on sow thistle. No *Alternaria* conidia were found on these leaves until specimens were held in damp chambers. Monoconidial isolates were obtained from sporulation produced under damp-chamber conditions.

### Materials and methods

For morphological observations cultures were obtained under conditions closely approximate to those recommended by E.G. Simmons (1992, 2007). Monoconidial isolates were cultivated in Petri dishes on potato-carrot agar (PCA) and V-4 (for 1 l medium: 150 ml juice mixture [beet, celery, carrot, tomato 4:3:2:1] and 20 g agar; Mikhailova et al., 2002), which is analogous to V-8, at 24°C under light/dark cycle (12/12 h). Preparations for microscopy

were made after 10–12 days of growth. All strains are kept in the All-Russian Institute of Plant Protection (St. Petersburg) and the All-Russian Collection of Microorganisms – VKM (Moscow). The dried leaves and dried cultures on PCA and V-4 of all strains are available at the herbarium of the Institute – LEP.

### Taxonomic description

*Alternaria silybi* Gannibal, sp. nov.

FIG. 1

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*Ex cultura in agar V-4 descripta. Conidiophora primaria solitaria, simplicia, ad ca. 50–90(–150) × 5.0–5.5 μm, apice dilatato ad 6.0–7.0 μm. Conidia solitaria; corpus conidiorum in maturitate longe anguste ellipsoideum vel subcylindricum, 50–90 × 15–22 μm, 5–10 transverse septatum, 1-longiseptatum in 1–4(–5) segmentis transversis, laeve, dilute brunneum, 1(–2)-rostratum. Rostrum filamentosum, 70–190 × 2.5–3.5 μm, 1–4(–5) transverse septatum. Habitatio typi in foliis vivis Silybum marianum, Russia, Primorskiy kray, Vladivostok, 1.IX.2006, leg. Ph. B. Gannibal.*

TYPE – Rusia. Primorskiy kray: Vladivostok, Trudovoe, Experimental and Industrial Farm 'Fruit and Berry Experimental Station' (43°18.18'N, 132°06.50'E), from leaf lesion of milk thistle, *Silybum marianum* (L.) Gaertn. (Asteraceae), 1.IX.2006, coll. Ph.B. Gannibal. (Holotype, LEP 12650 (dried V-4 agar culture); live strain, MF-P050011 (VKM F-4109)).

ETYMOLOGY: from the Latin *Silybum*, the host genus (milk thistle).

DESCRIPTION – On V-4 CULTURES are dark olive-grey, later almost black, velvety; AERIAL MYCELIUM is very weak or absent; diameter of 7-d old COLONIES is about 60 mm. On PCA COLONIES are almost colorless with pale brown or olive shade; AERIAL MYCELIUM is very weak or absent; diameter of 7-d old COLONIES is 25–35 mm.

On V-4 agar PRIMARY CONIDIOPHORES usually are solitary and uncrowded. They are simple with a single apical conidiogenous locus or sometimes with two loci; (35–)50–90(–150) × 5.0–5.5 μm swollen at the apex up to 6.0–7.0 μm. CONIDIA are solitary. In old cultures occasionally they can form CHAINS of 2 conidia.

JUVENILE CONIDIA are pale and wedge-shaped, long-narrow ellipsoid or subcylindric; usually they initiate production of a narrow-taper BEAK at a very early stage of development. The BODY of mature conidia is long-ellipsoid, subcylindric or long-ovoid; usually pale olive brown, sometimes dark; 50–80 × 15–20(–22) μm. Most conidial BODIES have (5–)7–10 TRANSVERSE SEPTA. LONGISEPTA may be absent or present as 1(–2) in 1–3 transverse segments, occasionally in 4–5 segments. The CONIDIAL BODY is slightly constricted near the transverse septa. Sometimes CONIDIA have specific shape of composite cylinder due to blocks of 1–3 transverse segments that have conspicuously different width in comparison with neighbor segments. Conidia have one

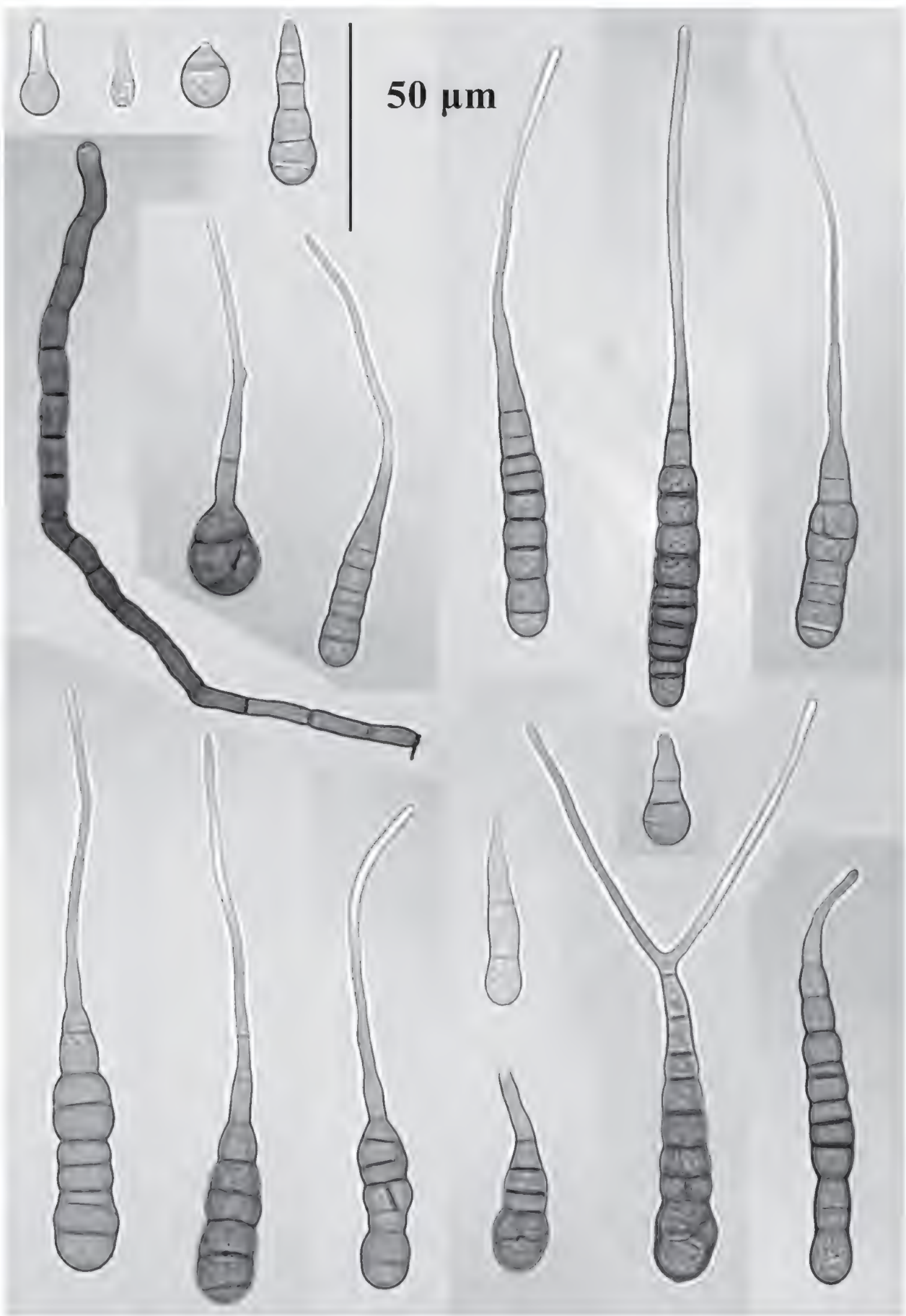


FIG. 1. *Alternaria silybi*: conidia and conidiophores ex holotype

BEAK, very rarely produce two beaks and/or apical and lateral SECONDARY CONIDIOPHORES. Filamentous BEAK length reaches into a range of 70–130 (–190)  $\mu\text{m}$ ; beaks are ca 3  $\mu\text{m}$  wide throughout most of their length and have 1–4 (–6) transverse septa. In most cases length of the BEAKS is the same as length of conidial body or rather more; rarely the beak is two times longer.

On PCA CONIDIA are negligibly bigger, 50–90  $\times$  15–22  $\mu\text{m}$  (body) + 100–190  $\mu\text{m}$  (beak).

STRAINS EXAMINED – RUSSIA. PRIMORSKIY KRAY: Vladivostok, Trudovoe, Experimental and Industrial Farm ‘Fruit and Berry Experimental Station’ (43°18.18’N, 132°06.50’E)—from leaf lesion of milk thistle, 1.IX.2006 (VKM F-4109 and F-4118).

PRIMORSKIY KRAY: Vladivostok, Botanical Garden-Institute—from leaf lesion of milk thistle, 6.IX.2006 (VKM F-4117).

COMMENTS – *A. silybi* is similar to *A. protenta* E.G. Simmons, which was also found on *Asteraceae*. *A. silybi* differs by smaller maximal conidium body size, longer beak lengths, and smooth walls.

***Alternaria simmonsii* Gannibal, sp. nov.**

FIG. 2

MYCOBANK MB518504

*Ex cultura in agaro V-4 descripta. Conidiophora primaria solitaria, simplicia, ad ca. 40–200  $\times$  5–6  $\mu\text{m}$ , brunnea. Conidia solitaria vel in catenis conidiorum bini. Corpus conidiorum late ovoideum vel ellipsoideum, ad 50–90  $\times$  22–30(–36)  $\mu\text{m}$ ; 5–8 transverse septatum, 1–3 longiseptatum, clare brunneum. Conidia rostrata vel erostrata, rostro longo ad 100  $\times$  3  $\mu\text{m}$ , 1–2(–4) transverse septata. Habitatio typi in foliis vivis *Sonchus* sp., Russia, Voronezhskaya oblast, Semilukskiy rayon, selo Veduga, 20.V.2005, leg. I. V. Bilder.*

TYPE – Russia. Voronezhskaya oblast: Semilukskiy rayon, selo Veduga, from leaf lesion of sow thistle, *Sonchus* sp. (*Asteraceae*), 20.V.2005, coll. I.V. Bilder. (Holotype, LEP 12651 (dried V-4 agar culture); live strain, MF-P024011 (VKM F-4110)).

ETYMOLOGY: the epithet honours Emory G. Simmons, who has studied *Alternaria* taxonomy for 50 years.

DESCRIPTION – On V-4 CULTURES are dark olive, later almost black, velvety; AERIAL MYCELIUM is sparse; diameter of 7-d old COLONIES is ca 65 mm. On PCA COLONIES are pale brown or light olive grey; AERIAL MYCELIUM is very weak or absent; diameter of 7-d old COLONIES is ca 40 mm.

PRIMARY CONIDIOPHORES on V-4 agar arise directly from the agar substrate surface or from branches of the woolly aerial mycelium. Usually they are solitary, simple, straight or slightly sinuous, 40–200  $\times$  5–6  $\mu\text{m}$ , with a single apical conidiogenous locus or sometimes with two loci. CONIDIA are solitary; sometimes they can form chains of 2 conidia.

JUVENILE CONIDIA are ovate, rarely ellipsoid or cylindrical, light brown, commonly without beak. The MATURE CONIDIUM BODY is brown, long ovoid, ellipsoid or bag-shaped, sometimes asymmetric, and becomes fully developed in a size range of ca 50–90  $\times$  22–30(–36)  $\mu\text{m}$ . It has 5–8 main transverse



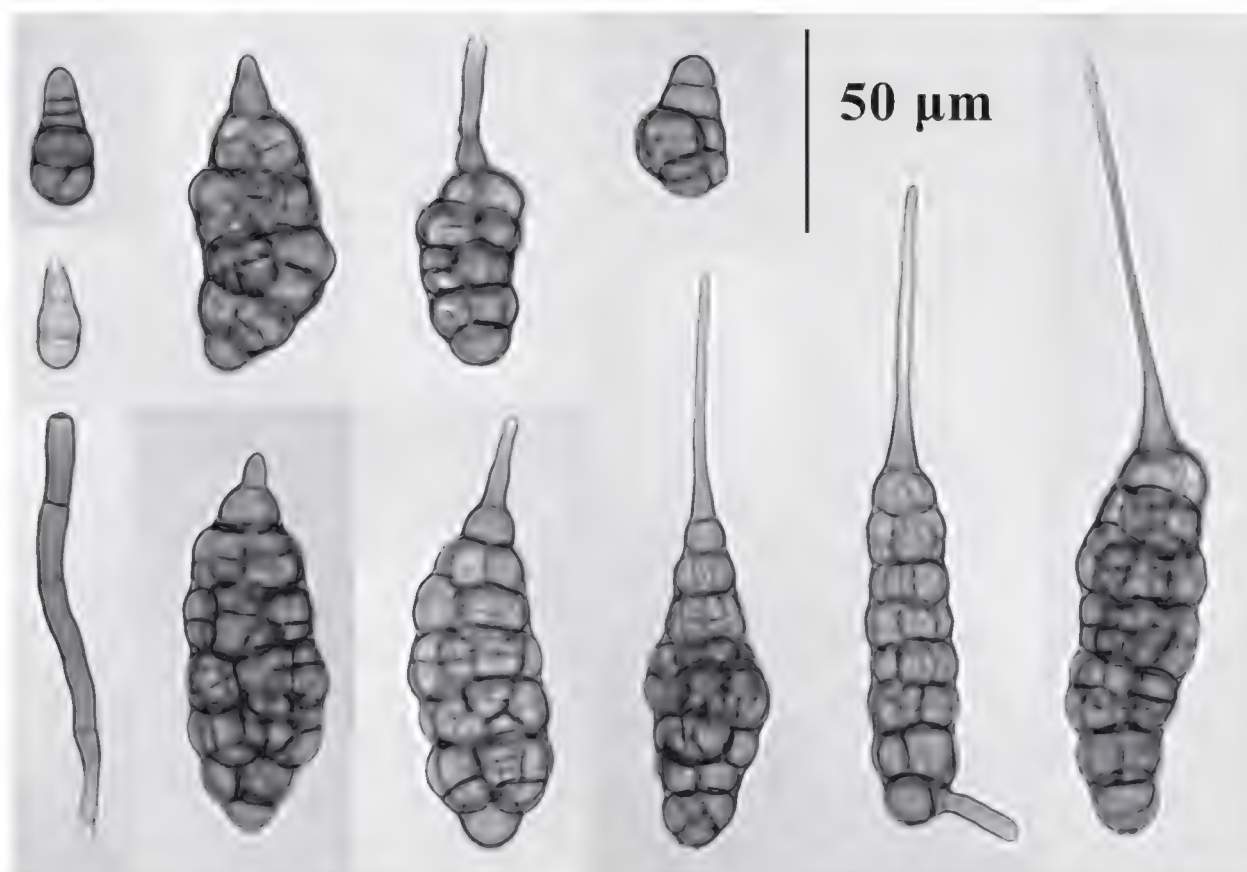


FIG. 2. *Alternaria simmonsii*: conidia and conidiophores ex holotype

constricting divisions. Nearly all well-developed conidia have 1–3 longitudinal and 1 secondary transverse DISTOSEPTUM divisions in most of the transverse segments. The filamentous, unbranched solitary BEAK frequently is lacking; when present it is variable in length, and sometimes becomes as long as 100  $\mu\text{m}$ . BEAKS are ca 3  $\mu\text{m}$  wide throughout most of their length and have 1–2(–4) transverse septa. Sometimes BEAKS are slightly swollen at the end. Some basal conidia form 1 apical or/and 1–2 lateral SECONDARY CONIDIOPHORES.

On PCA the CONIDIAL BODY has a more regular ellipsoid shape than on V-4 and is smaller ( $40\text{--}75 \times 17\text{--}23 \mu\text{m}$ ); however, the BEAK is conspicuously longer and sometimes reaches 155  $\mu\text{m}$  long.

STRAINS EXAMINED – RUSSIA. VORONEZHSKAYA OBLAST: Semilukskiy rayon, selo Veduga—from leaf lesion of sow thistle, 20.V.2005 (VKM F-4110 and F-4119).

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**The *Entolomataceae* of the Pakaraima Mountains of Guyana 5:  
new species of *Alboleptonia***T.W. HENKEL<sup>\*1</sup>, M.C. AIME<sup>2</sup>, D.L. LARGENT<sup>1</sup> & T.J. BARONI<sup>3</sup><sup>\*</sup> *tw5@humboldt.edu*<sup>1</sup>*Department of Biological Sciences, Humboldt State University  
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**Abstract**—This paper is the fifth in a series documenting the *Entolomataceae* taxa (*Agaricales*, *Basidiomycota*) from Guyana. Three new species are described — *Alboleptonia angustospora*, *A. cystidiosa*, and *A. minima* — occurring in tropical rainforests of the Upper Potaro River Basin in Guyana's Pakaraima Mountains. Macromorphological, micromorphological, and habitat data are provided for each. *Alboleptonia* has not been previously reported from Guyana.

**Key words**—*Agaricomycotina*, fungal taxonomy, Guayana Highlands, Guiana Shield, neotropics

**Introduction**

Species of *Alboleptonia* Largent & R.G. Benedict are easily classified into the *Entolomataceae* (*Agaricales*) due to their dull pink basidiospores that are angular in all views. *Alboleptonia* was erected (Largent & Benedict 1970) to accommodate entolomatoid species that combine diagnostic features of the type species, *Alboleptonia sericella* (Fr.) Largent & R.G. Benedict, including a white to pale cinereous basidioma, a silky to appressed-fibrillose or minutely appressed-squamulose, opaque (NOT translucent striate), non-hygrophanous pileal surface, which microscopically is composed of an entangled layer of hyphae, unique color reactions in Ehrlich's reagent, and a low urea concentration. Also, under scanning electron microscopy *Alboleptonia* basidiospores exhibit a dihedral base and a pair of 4-angled facets on the apico-adaxial side that

results in a 5-sided apical facet (Pegler & Young, 1978). This original concept of *Alboleptonia* has subsequently been applied by Largent (1994), Baroni & Lodge (1998), Pegler (1983, 1997), and Orton (1991a, b). A recent molecular study (Co-David et al. 2009), which putatively shows *Alboleptonia* as polyphyletic, suffered from small sample size (two species) and incongruent application of generic/subgeneric concepts regarding *Alboleptonia* sensu Largent & Benedict and *Entoloma* subgen. *Alboleptonia* (Largent & R.G. Benedict) Noordel. (Noordeloos 1979, 1987, 1988, 1992, 2004).

New World tropical and subtropical species meeting the diagnostic requirements of *Alboleptonia* sensu Largent & Benedict have been found in the Lesser and Greater Antilles (Baroni & Lodge 1998; Pegler 1983), Trinidad and Venezuela (Dennis 1953, 1970), Brazil (Pegler 1997), and elsewhere in South America (Horak 1977, 1982). Over the course of several years of field expeditions in a remote region of the Pakaraima Mountains of Guyana, we have collected fungi representing at least four distinct entolomatoid taxa corresponding to *Alboleptonia* sensu Largent & Benedict, three of which are described here.

### Materials and methods

Collections were made during the 2001–03, 2006, and 2009 May–July rainy seasons and the 2003 and 2009 December rainy seasons from the Upper Potaro River Basin, within a 15 km radius of a permanent base camp at 5°18'04.8"N; 59°54'40.4"W; elevation 710 m. This collecting area, located in an undulating valley approximately 20 km east of Mt. Ayanganna (2200 m), is densely forested with a mosaic of primary *Dicymbe*-dominated and mixed forests of the *Eschweilera*–*Licania* association (Henkel 2003). Methods for field descriptions, microscopic analyses, and image capture were those of Largent et al. (2008). Fungi were field-dried with silica gel. Color designations follow Kornerup & Wanscher (1978) with color plates noted in parentheses (e.g., 4A7). Specimens were deposited in the following herbaria: BRG, HSU, and LSUM (Holmgren et al. 1990). Microscopic structures were measured as described in Largent (1994) and Largent et al. (2008a). Statistics determined include: means of basidiospore length and width,  $\pm$  standard deviations; E, the quotient of length by width indicated as a range variation in n objects measured; Q, the mean of E-values; n = number of objects measured.

### Taxonomy

*Alboleptonia angustospora* Largent, Aime & T.W. Henkel, sp. nov.

FIG 1

MYCOBANK MB 518326

*Pileus* 10–17 mm *latus*, late *convexus* vel *plano-convexus*, ad *centrum depressus*, *albus* vel *eburneus*, *implexus* *appressus* *fibrillosus*, *siccus*. *Lamellae* *adnatae*, *sub-adnexae*, vel *subdecurrentes*, *subdistantes*, *albae* vel *roseae*; *marginae* *concolori*, *cystidiata*. *Stipes* 33–57

$\times 1\text{--}3\text{ mm}$ , *equalis*, *albus*, *glaber*, *apice pruinoso*. *Basidiosporae* 5–6-angulares,  $7.3\text{--}10 \times 5.1\text{--}7.6\text{ }\mu\text{m}$ . *Basidia* 4-sterigmata, late cylindracea,  $20\text{--}34.3 \times 6.6\text{--}10.4\text{ }\mu\text{m}$ . *Cheilocystidia* abundantes, cylindro-clavata. *Pleurocystidia* carentes. *Pileipellis* constata e intricatis hyphis. *Fibulae* carentes.

TYPE: Aime 3159 (BRG, holotype; LSUM, isotype).

ETYMOLOGY: *angustus* (L. adj.) = narrow; *-sporus* (L. adj.) = spored; referring to the narrow basidiospores.

KEY CHARACTERS — *Alboleptonia angustospora* is easily recognized as a member of *Alboleptonia* because of its white, non-hygrophanous, non-striate, convex-depressed (occasionally umbonate), entirely matted-tomentulose to matted-fibrillose pileus and its 5–6-angled, heterodiametric basidiospores. It is unique among macromorphologically similar species of *Alboleptonia* in its combination of cylindric to cylindro-clavate, somewhat strangulated cheilocystidia, 5–6-angled, heterodiametric basidiospores that average  $< 9\text{ }\mu\text{m}$  long and  $< 7\text{ }\mu\text{m}$  broad, and the lack of pleurocystidia, clamp connections, and pigmentation.

MACROCHARACTERS — PILEUS 10–17 mm broad, 5–8 mm high, broadly convex to plano-convex with a distinct central depression occasionally with a very small, blunt umbo, entirely matted-tomentulose to matted-appressed fibrillose, chalky white to off-white to pale cream (4A1–4A2) at times with a faint hint of yellow (2A4) at disc, opaque, dry, not hygrophanous, not translucent; margin somewhat downcurved, entire but under hand lens irregularly and finely crenulate. LAMELLAE subclose to subdistant, adnate, subadnexed, or subdecurrent, 1.5–2.4 mm tall, chalky white, faintly pink at maturity (5A2–5A3); margin concolorous, finely eroded-cystidiate under hand lens; lamellulae 3, of different lengths. STIPE 33–57 mm  $\times$  1–3 mm, equal, glabrous, occasionally white-pruinose at apex, concolorous, yellowing with age, cartilaginous, very fragile, hollow. BASAL MYCELIUM scant, white. ODOR none, pleasantly fungoid, or slightly fragrant; TASTE slightly fungoid. SPORE DEPOSIT not obtained.

MICROCHARACTERS — BASIDIOSPORES distinctly 5–6-angled, isodiametric in polar view, subisodiametric to heterodiametric (rarely isodiametric) in profile view,  $7.3\text{--}10 \times 5.1\text{--}7.6\text{ }\mu\text{m}$  (mean =  $8.5 \pm 0.56 \times 6.44 \pm 0.54\text{ }\mu\text{m}$ ;  $E = 1.1\text{--}1.68$ ,  $Q = 1.33 \pm 0.12$ ,  $n = 104$ ). BASIDIA 4-sterigmate, broadly cylindric and rounded at the base,  $20\text{--}34.3\text{ (–}38.4) \times 6.6\text{--}10.4\text{ }\mu\text{m}$  (mean =  $28.0 \pm 2.9 \times 8.8 \pm 0.79\text{ }\mu\text{m}$ ;  $E = 2.3\text{--}4.2$ ,  $Q = 3.1 \pm 0.49$ ;  $n = 29$ ). CHEILOCYSTIDIA abundant, cylindric to cylindro-clavate, many somewhat strangulated,  $17.3\text{--}86.0 \times 3.8\text{--}9.4\text{ }\mu\text{m}$  (mean =  $46.8 \pm 15.94 \times 6.0 \pm 1.28\text{ }\mu\text{m}$ ;  $E = 2.15\text{--}19.13$ ,  $Q = 7.72$ ;  $n = 29$ ). PLEUROCYSTIDIA absent. LAMELLAR TRAMA composed of parallel to subparallel, rather short hyphae, cells  $44.8\text{--}145.1 \times 2.4\text{--}15.9\text{ }\mu\text{m}$ . PILEIPELLIS an entangled layer of hyphae throughout; terminal cells cylindric to cylindro-clavate,  $23.4\text{--}57.1 \times 5.6\text{--}11.3\text{ }\mu\text{m}$ . PILEAL TRAMA composed of entangled hyphae, cells  $44.3\text{--}140.2$



$\times 6.3\text{--}21.0\ \mu\text{m}$ . STIPITIPPELLIS a cutis; hymenial clusters occasionally present; caulocystidioid elements  $45.0\text{--}55.4 \times 2.4\text{--}6.7\ \mu\text{m}$ . REFRACTIVE HYPHAE scattered to abundant in the pileal trama. REFRACTIVE GRANULES, BRILLIANT GRANULES, and PIGMENTATION absent. CLAMP CONNECTIONS absent.

ECOLOGY, RANGE, DISTRIBUTION — Solitary on humic mat on forest floor or clay soil in mixed *Dicymbe* spp. forest, known only from the Upper Potaro River Basin of Guyana.

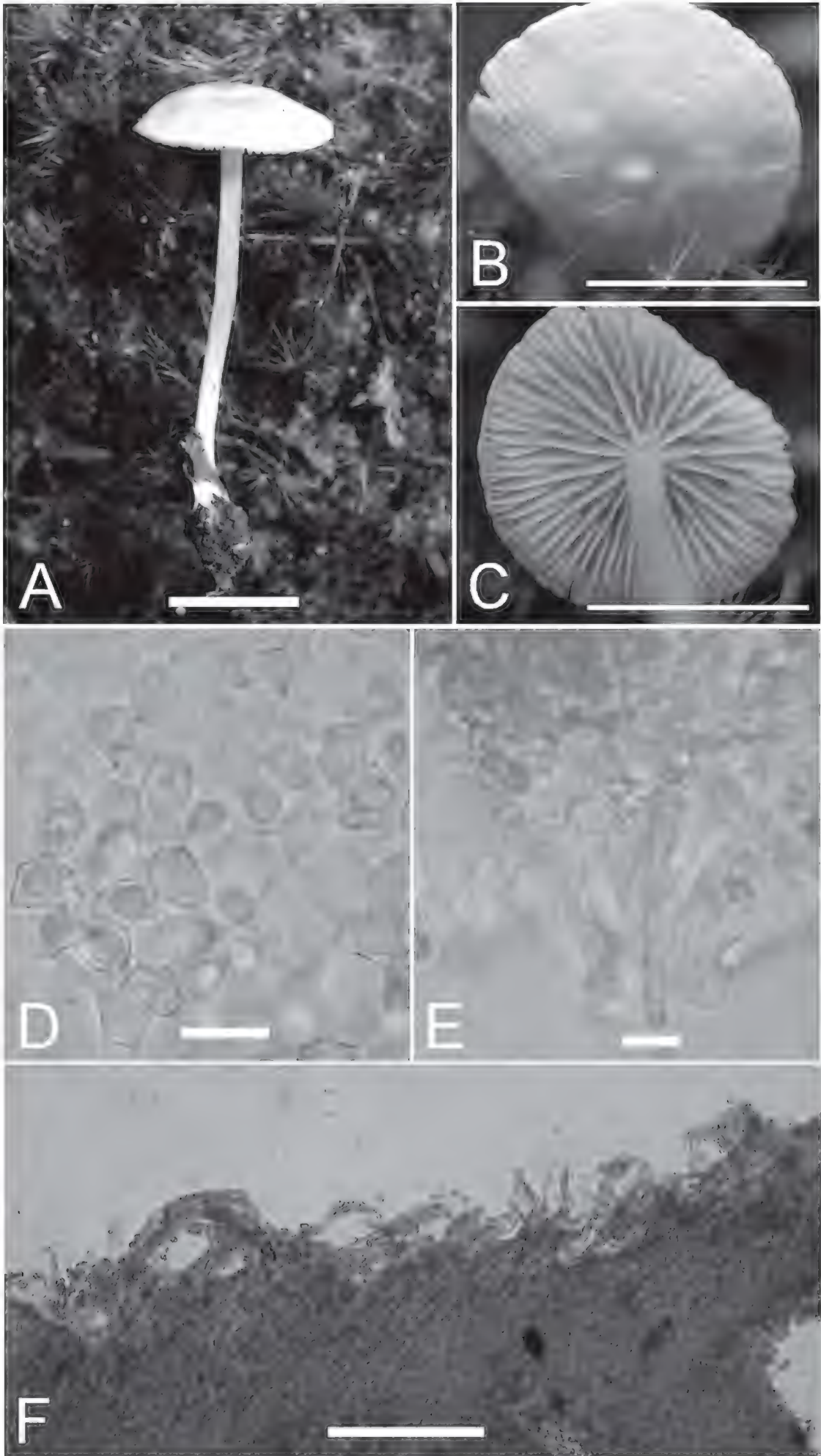
REPRESENTATIVE SPECIMENS EXAMINED. GUYANA. REGION 8: POTARO-SIPARUNI. Pakaraima Mountains. Upper Potaro River Basin, 15–20 km east of Mt. Ayanganna, environs of base camp located on Potaro River one km upstream from confluence with Whitewater Creek at  $5^{\circ}18'04.8''\text{N}$ ,  $59^{\circ}54'40.4''\text{W}$ , elevation 710–750 m: vicinity of base camp, 11 May 2001, *Henkel* 8095 (BRG; HSU); 2.5 km southeast of base camp, *Dicymbe* plot 2 in humic mat, 12 June 2002, *Aime* 1978 (BRG; LSUM); 0.5 km west of base camp, in *Dicymbe* forest, 29 June 2002, *Aime* 2161 (BRG; LSUM); 1 km southeast of base camp on Benny's ridge in clay soil, 2 July 2006, *Aime* 3159 (BRG, holotype; LSUM, isotype); vicinity of Tadang Creek base camp, 29 December 2009, *Henkel* 9148 (BRG; HSU).

COMMENTS — *Alboleptonia angustospora* resembles a group of species including *Entoloma parasericellum* Corner & E. Horak, *E. neosericellum* E. Horak, *E. subsericellum* Murrill, *E. peralbidum* Horak, *E. percandidum* Noordel., and *E. hololeucum* (Singer) E. Horak. *Alboleptonia angustospora* can be separated from this group of species by a combination of the following characters: cylindric to cylindro-clavate cheilocystidia, 5–6-angled, heterodiametric basidiospores that average  $< 9\ \mu\text{m}$  in length and  $< 7\ \mu\text{m}$  in width, and the lack of pleurocystidia, clamp connections, and pigmentation.

In Guyana, *A. angustospora* may be confused with *Alboleptonia minima* and *A. cystidiosa* (described here) as each of these species has a white basidioma with an appressed-fibrillose, opaque, non-translucent striate pileus, similarly shaped and sized basidiospores, and lacks clamp connections. *Alboleptonia minima* can be separated from *A. angustospora* by its small pileus ( $< 10\ \text{mm}$  broad) and somewhat longer stipe (both of which lack cream or yellowish tones), dense tomentose basal mycelium, and anatomically similar stipitipellis, pileipellis, and lamellar edges that include non-strangulated cheilocystidia. *Alboleptonia cystidiosa* is distinct from *A. angustospora* due to its cylindro-clavate caulocystidia, clavate to obclavate cheilocystidia and pleurocystidia, and weakly acrid taste. In Guyana, several other as yet unidentified white entolomatoid species superficially resemble *A. angustospora*. However these taxa either have differently shaped basidiospores and/or a different pileipellis structure compared to *A. angustospora* (Henkel & Aime unpubl. data).

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FIG. 1. Macro- and microscopic features of *Alboleptonia angustospora* (BRG HOLOTYPE *Aime* 3159). A. Basidioma. B. Matted-fibrillose pileus surface with umbo. C. Lamellae with cystidiate margins. Bar = 10 mm. D. Basidiospores. E. Cheilocystidia. Bar =  $10\ \mu\text{m}$ . F. Pileus surface in longitudinal section. Bar =  $100\ \mu\text{m}$ .



*Alboleptonia earlei* (Murrill) Largent & R.G. Benedict from Cuba and Costa Rica (Baroni & Lodge, 1998) is the only other neotropical *Alboleptonia* species known that lacks clamp connections and has basidiospores similar in size ( $7\text{--}9 \times 5.5\text{--}6.5 \mu\text{m}$ ) and shape to those of *A. angustospora*. *Alboleptonia earlei* can be differentiated by its lack of cheilocystidia and garlic or onion odor (Largent & Benedict 1970; Baroni & Lodge 1998).

Among Old World alboleptonioid fungi, *Entoloma inficetum* Corner & E. Horak from the Solomon Islands has many of the same characteristics as *Alboleptonia angustospora*. However, *E. inficetum* has a smooth pileus with an entirely repent pileipellis and cheilocystidia with a yellowish, protoplasmic pigment; in *A. angustospora*, the pileus is consistently matted-fibrillose to matted-tomentose, the pileipellis is an entangled hyphal layer that is never repent, and the cheilocystidia lack pigment (Horak 1980).

***Alboleptonia cystidiosa* Largent & Aime, sp. nov.**

FIG 2

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*Pileus 8–35 mm latus, convexus vel plano-convexus, albus, cinereo humili umbone centrum occupanti, radiatim appressus fibrillosus. Lamellae adnatae, concolores. Stipes 25–48 × 2.5–7 mm, concolor. Basidiosporae 5–6-angulares, 7.6–9.8 × 5.3–8.4 μm. Basidia 2 vel 4-sterigmatae, clavatae, 28–38.2 × 7.6–10.7 μm. Cheilocystidia et pleurocystidia abundantes, obclavata. Pileipellis constata e intricatis hyphis. Fibulae carentes.*

TYPE: Aime 2395 (BRG, holotype; LSUM, isotype).

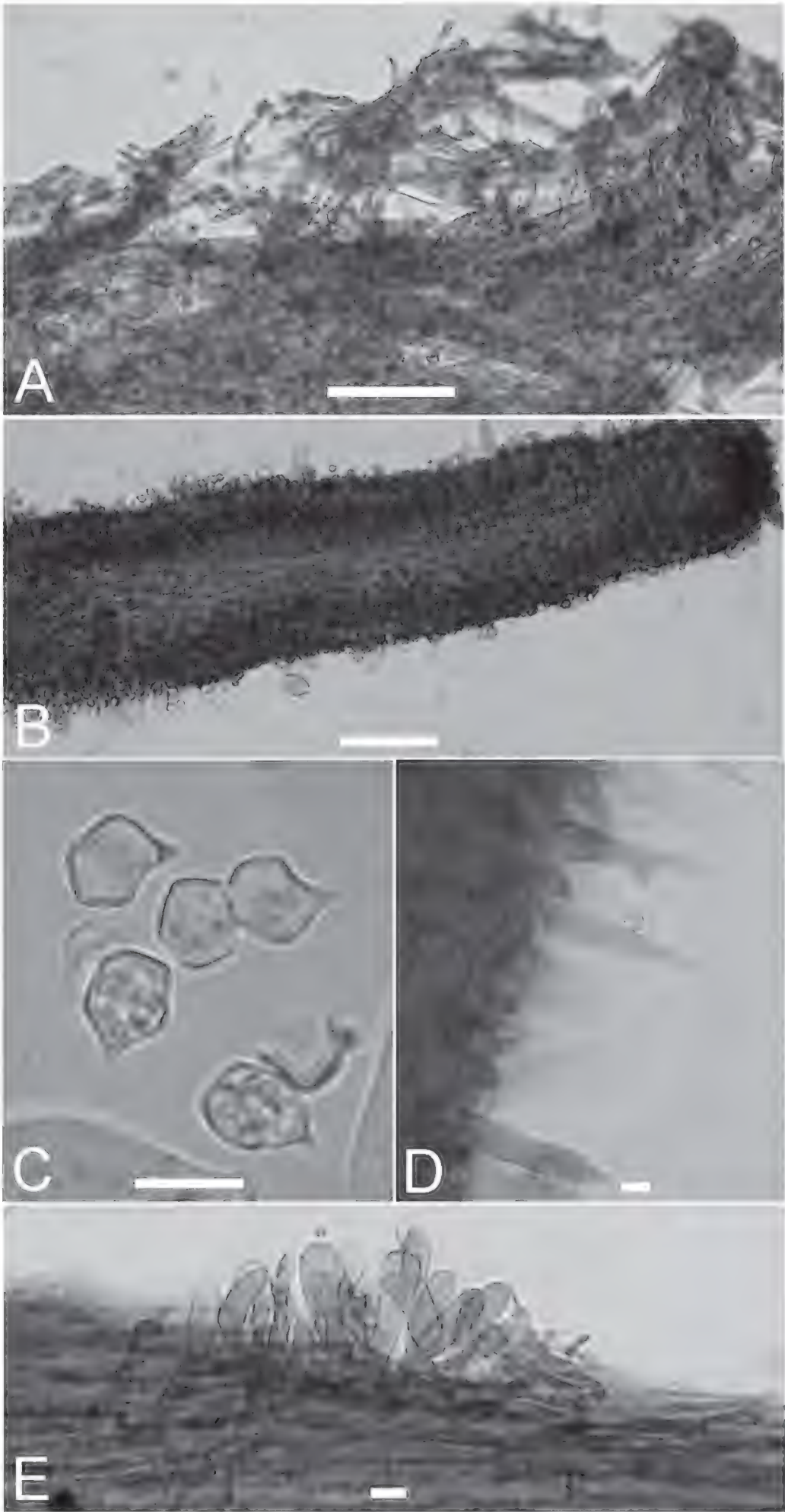
ETYMOLOGY: *cystidiosus* (L. adj.) referring to the abundant hymenial cystidia.

**KEY CHARACTERS** — *Alboleptonia cystidiosa* is unique in its combination of a convex pileus with a rounded to flattened grayish umbo, slightly acrid taste, small, heterodiametric basidiospores, abundant clavate to obclavate cheilocystidia and pleurocystidia, cylindro-clavate to clavate caulocystidia, and lack of clamp connections.

**MACROCHARACTERS** — **PILEUS** 8–35 mm broad, narrowly convex to broadly convex to nearly plane but wavy with age, chalky white with a greyish, low flattened umbo; appearing glabrous, under hand lens radially fibrillose, scurfy over umbo; margin entire, finely eroded with age; trama very thin, < 1 mm over stipe. **LAMELLAE** close, adnate, thin, narrow, < 1 mm tall, white, faintly pink at maturity, occasionally forking near margin; lamellulae 4–6, of different lengths. **STIPE** 25–48 × 2.5–7.0 mm, slightly broader and flattened towards base, chalky white, glabrous, finely longitudinally striate under handlens; context white, unchanging, hollow. **BASAL MYCELIUM** lacking. **ODOR** faint, indistinct; **TASTE** slightly acrid. **SPORE DEPOSIT** salmon pink (7B4).

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FIG. 2. Microscopic features of *Alboleptonia cystidiosa* (BRG HOLOTYPE Aime 2395). A. Pileus surface in longitudinal section. B. Lamellar section showing abundant pleurocystidia. Bar = 100 μm. C. Basidiospores. D. Cheilocystidia. E. Caulocystidia near stipe apex. Bar = 10 μm.





**MICROCHARACTERS** — **BASIDIOSPORES** distinctly 5–6-angled, isodiametric in polar view, subisodiametric to heterodiametric in profile view, rarely isodiametric,  $7.6\text{--}9.8 \times 5.3\text{--}8.4\ \mu\text{m}$ , (mean =  $8.88 \pm 0.53 \times 6.92 \pm 0.72\ \mu\text{m}$ ,  $E = 1.09\text{--}1.56$ ,  $Q = 1.29 \pm 0.1$ ;  $n = 28$ ). **BASIDIA** clavate, 2 or 4-sterigmate,  $28.0\text{--}38.2 \times 7.6\text{--}10.7\ \mu\text{m}$ , ( $E = 3.07\text{--}4.72$ ,  $Q = 3.7 \pm 0.38$ ;  $n = 13$ ). **CHEILOCYSTIDIA** abundant, obclavate, occasionally clavate, hyaline,  $36.0\text{--}129.0 \times 8.8\text{--}15.4\ \mu\text{m}$ . **PLEUROCYSTIDIA** abundant, similar in shape to but smaller than the cheilocystidia, hyaline,  $47.2\text{--}86.6 \times 10.0\text{--}15.84\ \mu\text{m}$ . **PILEIPELLIS** an entangled layer of cylindric hyphae throughout. **PILEOCYSTIDIA** clavate to cylindro-clavate,  $20.5\text{--}50.9 \times 4.5\text{--}15.7\ \mu\text{m}$ . **CAULOCYSTIDIA** in scattered but abundant clusters, cylindro-clavate to clavate to broadly clavate,  $17.3\text{--}59.1 \times 5.3\text{--}19.3\ \mu\text{m}$ . **REFRACTIVE HYPHAE** scattered in the pileus and stipe tramas. **REFRACTIVE GRANULES**, **BRILLIANT GRANULES**, and **PIGMENTATION** absent. **CLAMP CONNECTIONS** absent.

**ECOLOGY, RANGE, DISTRIBUTION** — Clustered on sandy soils in mixed riverine forest, known only from the Upper Potaro River Basin of Guyana.

**REPRESENTATIVE SPECIMENS EXAMINED.** **GUYANA.** **REGION 8:** POTARO-SIPARUNI. Pakaraima Mountains. Upper Potaro River Basin, ~15 km east of Mt. Ayanganna, environs of Ayanganna airstrip, elevation ~720 m: on trail between airstrip and Potaro River in sandy soil, 29 December 2003, *Aime* 2395 (BRG, holotype; LSUM, isotype).

**COMMENTS** — *Alboleptonia cystidiosa* is similar to the pantropical *Alboleptonia stylophora* (Berk. & Broome) Pegler and *Entoloma niveum* G. Stev. from New Zealand in possessing a white, umbonate pileus that is non-hygrophanous and non-striate, cheilocystidia, and an absence of clamp connections. *Alboleptonia stylophora* can be distinguished from *A. cystidiosa* by its cuspidate pileus that tends to develop yellowish hues, cylindro-clavate cheilocystidia, lack of pleurocystidia, and considerably larger basidiospores ( $9.3\text{--}13.8 \times 7.7\text{--}9.7\ \mu\text{m}$ ; Baroni & Lodge, 1998). *Entoloma niveum* differs from *A. cystidiosa* in its papillate pileus, farinaceous odor, strangulated cheilocystidia, and lack of pleurocystidia (Horak 1973, 2008). *Entoloma neoseriellum* from New Zealand resembles *A. cystidiosa* in having a white, innately fibrillose pileus, cheilocystidia and pleurocystidia, and an absence of clamp connections. *Entoloma neoseriellum* is nonetheless easily separated from *A. cystidiosa* by its ventricose-rostrate pleurocystidia, larger basidiospores ( $10\text{--}11.5 \times 7.5\text{--}8.5\ \mu\text{m}$ ), and hygrophanous, translucent-striate, non-umbonate pileus (Horak 2008).

*Alboleptonia minima* Largent & T.W. Henkel, sp. nov.

FIG 3

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*Pileus* 7–8 mm *latus*, late *convexus* vel *planus*, ad *centrum depressus*, *albus*, minute *appressus-fibrillosus*. *Lamellae* *adnatae*, *confertae*, *albae* vel *roseae*. *Stipes* 50–56  $\times$  2–3 mm, *apicem* versus *leviter contractus*, *albus*, *apice pruinosis*. *Basidiosporae* 6-angulares,  $7.5\text{--}9.1 \times 5.8\text{--}7.4\ \mu\text{m}$ . *Basidia* 4-sterigmata, *clavata*,  $24.3\text{--}31.7 \times 7.2\text{--}9.5\ \mu\text{m}$ . *Cheilocystidia* abundantes, *cylindro-clavata*. *Pleurocystidia* carentes. *Pileipellis* *constata* e intricatis *hyphis sub-erectis terminalibus cellulis*. *Fibulae* carentes.



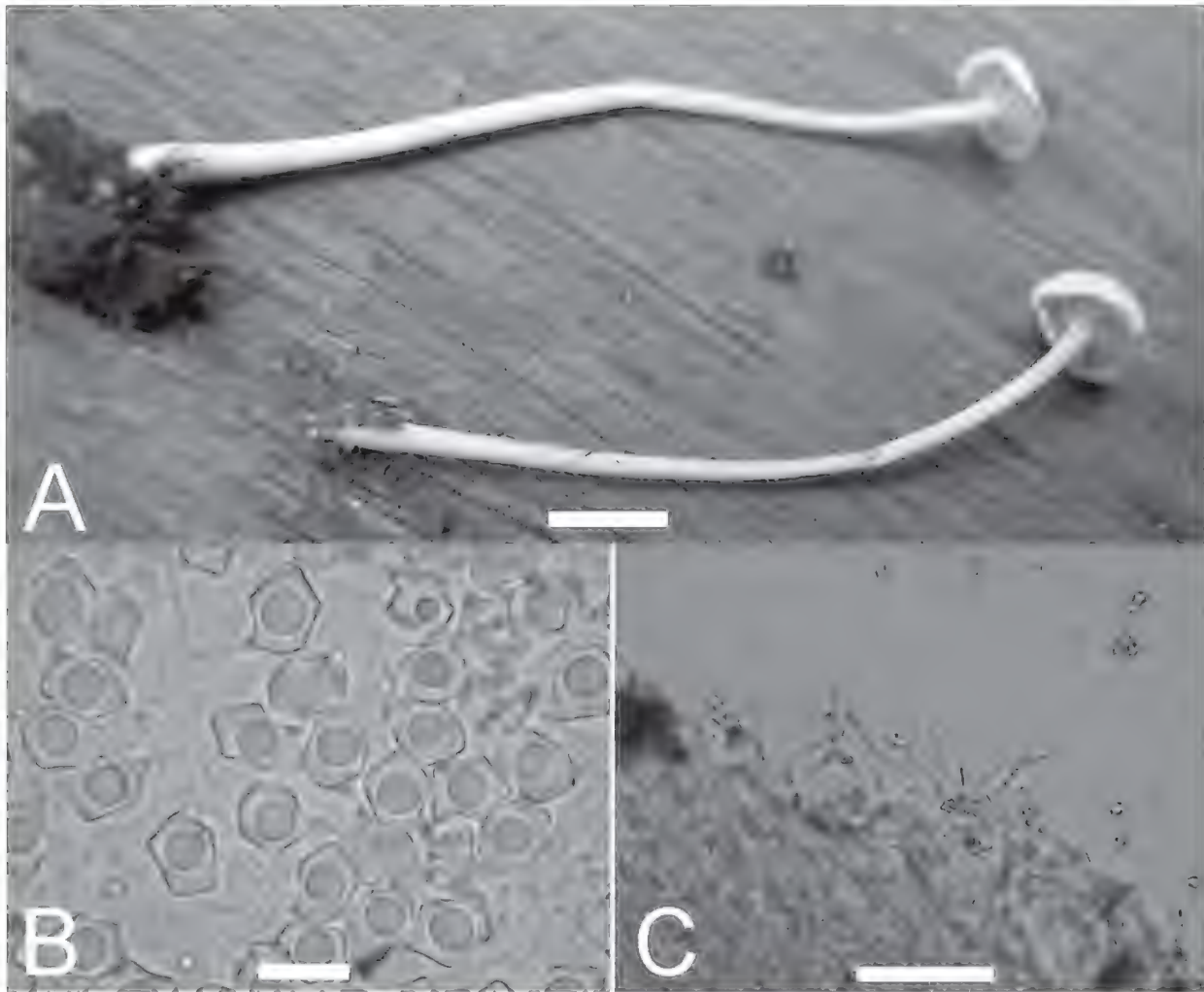


FIG. 3. Macro- and microscopic features of *Alboleptonia minima* (BRG HOLOTYPE Henkel 9037).  
A. Basidiomata. Bar = 10 mm. B. Basidiospores. C. Pileipellis with sub-erect terminal elements.  
Bar = 10  $\mu$ m.

TYPE: *Henkel 9037* (BRG, holotype; HSU, isotype).

ETYMOLOGY: *minimus* (L. adj.) = very small or tiny, referring to the width of the pileus.

KEY CHARACTERS — *Alboleptonia minima* is distinguished by its white basidioma with a depressed < 10 mm broad pileus, narrow, relatively long stipe, lack of clamp connections, and a stipitipellis, pileipellis, and lamellar edges composed of an entangled layer of hyphae.

MACROCHARACTERS — PILEUS 7–8 mm broad, 1–2 mm high, broadly convex to plane with a broad central depression, white, minutely appressed-fibrillose, opaque, not translucent, not hygrophanous; margin decurved to nearly plane, entire. LAMELLAE 2–3 mm long, 1 mm tall, white at first, faintly pink with age, adnate, close; margin minutely fringed under hand lens; lamellulae not recorded. STIPE 50–56  $\times$  2–3 mm, enlarging slightly toward base, white, minutely pruinose at the apex and minutely fibrillose elsewhere, hollow. BASAL MYCELIUM a moderately dense white tomentum. TASTE, ODOR, and SPORE DEPOSIT not recorded.

**MICROCHARACTERS** — **BASIDIOSPORES** distinctly 6-angled, isodiametric in polar view, subisodiametric or more often heterodiametric in profile view, apex typically rounded and triangular,  $7.5\text{--}9.1 \times 5.8\text{--}7.4 \mu\text{m}$  (mean =  $8.5 \pm 0.4 \times 6.6 \pm 0.5 \mu\text{m}$ ;  $E = 1.15\text{--}1.47$ ,  $Q = 1.29 \pm 0.1$ ;  $n = 28$ ). **BASIDIA** 4-sterigmate, clavate, distinctly tapered downward,  $24.3\text{--}31.7 \times 7.2\text{--}9.5 \mu\text{m}$  (mean =  $28.1 \pm 2.4 \times 8.61 \pm 0.6 \mu\text{m}$ ;  $E = 2.66\text{--}3.95$ ;  $Q = 3.27 \pm 0.3$ ;  $n = 14$ ). **LAMELLAR EDGE** a sterile layer of entangled hyphae. **CHEILOCYSTIDIA** abundant, cylindric to cylindro-clavate,  $31.1\text{--}47.6 \times 3.8\text{--}5.5 \mu\text{m}$ . **PLEUROCYSTIDIA** absent. **LAMELLAR TRAMA** subparallel, of relatively short and narrow hyphae, cells  $48.9\text{--}87.5 \times 3.0\text{--}4.3 \mu\text{m}$ . **PILEIPELLIS** an entangled layer of hyphae with semi-erect terminal cells, particularly over disc. **PILEOCYSTIDIA** cylindric to narrowly cylindro-clavate,  $21.5\text{--}38.9 \times 2.8\text{--}8.3 \mu\text{m}$ . **PILEUS TRAMA** composed of interwoven hyphae, cells  $68.0\text{--}110.9 \times 7.0\text{--}10.4 \mu\text{m}$ . **STIPITIPELLIS** an entangled hyphal layer. **CAULOCYSTIDIA** similar in size and shape to the cheilocystidia. **REFRACTIVE HYPHAE** abundant in the subhymenium and pileus trama adjacent to lamellae, yellowish in 3% KOH, apparently absent in the lamellar trama. **REFRACTIVE GRANULES**, **BRILLIANT GRANULES**, and **PIGMENTATION** absent. **CLAMP CONNECTIONS** absent.

**ECOLOGY, RANGE, DISTRIBUTION** — Scattered on humus of forest floor in *Dicymbe* forest, known only from the Upper Potaro River Basin of Guyana.

**REPRESENTATIVE SPECIMENS EXAMINED.** GUYANA. REGION 8: POTARO-SIPARUNI. Pakaraima Mountains. Upper Potaro River Basin, 15–20 km east of Mt. Ayanganna, environs of base camp located on Potaro River one km upstream from confluence with Whitewater Creek at  $5^{\circ}18'04.8''\text{N}$ ,  $59^{\circ}54'40.4''\text{W}$ , elevation 710–750 m: in *Dicymbe* plot 2, 11 July 2009, Henkel 9037 (BRG, holotype; HSU, isotype).

**COMMENTS** — *Alboleptonia minima* is unique among entolomatoid fungi worldwide because of its white basidioma with a depressed pileus < 10 mm broad, narrow, relatively long stipe, and a stipitipellis, pileipellis, and lamellar edge composed of a similarly entangled layer of hyphae. Although *Rhodophyllus pilosellus* Romagn. & Gilles from Gabon shares a number of features with *A. minima*, it can be differentiated by its strongly fibrillose to flocculose pileus and its broader (11–17  $\mu\text{m}$ ) cheilocystidia that are covered over their apices with a hyaline, resinous substance (Romagnesi & Gilles 1979).

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## MYCOTAXON

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***Tuber foetidum* found in Finland**

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**Abstract** – *Tuber foetidum*, a white truffle belonging to the *T. macrosporum* group, is confirmed from Finland based on morphological and DNA analyses. The Finnish specimen was found in soil with relatively high pH in coniferous forest. The phylogenetic tree based on nuclear ribosomal ITS sequences indicated that the Finnish material is most similar to, but not identical with, *Tuber foetidum* samples from Hungary and Estonia.

**Key words** – Ascomycota, Tuberales, ectomycorrhiza, nrITS sequence

**Introduction**

Truffles, as strictly defined, are hypogeous fungi of the genus *Tuber*, which grow in symbiosis with certain trees. Due to rather controversial taxonomic treatments of large numbers of synonyms and varying species definitions, the real number of species is still unknown. The genus is mainly distributed in the Northern Hemisphere (Jeandroz et al. 2008). Truffles in Fenno-Scandinavia are less well documented compared with the Mediterranean region. Fries (1909), who gave the first modern account of *Tuber* species in Scandinavia, listed three species: *T. aestivum* Vittad., *T. maculatum* Vittad., and *T. rufum* Picco. Up to now, Denmark has the most records in this region, with 6 white and 3 black truffle species (Lange 1956). Five *Tuber* species are known from Sweden, including two black truffles, *T. aestivum* and *T. mesentericum* Vittad. (Danell 1996, Wedén et al. 2001). Recently the Burgundy truffle (*T. aestivum* f. *uncinatum* (Chatin) Montecchi & Borelli) has been produced on a small commercial scale in Gotland (Wedén et al. 2009). In Finland, where truffles are



not part of the culinary tradition, the first records of *Tuber* are *T. borchii* Vittad. and *T. maculatum* (Kosonen 2002). *Tuber borchii* is the only truffle species with gastronomic value found in Finland so far.

On 26 November 2006 a truffle ascocarp was found in a natural spruce forest dominated by *Picea abies* trees located in Lahti, Finland (100 km north to Helsinki) with the help of Ciro, a trained truffle dog. The truffle was morphologically and molecularly confirmed as *T. foetidum* Vittad., which represents the third *Tuber* species in Finland and the northernmost record for the species.

## Materials and methods

### Morphology

Morphological examinations of the ascocarp followed methods set forth in Pegler et al. (1993). Macroscopical descriptions are based on the field notes of the fresh ascocarp. The collection was air-dried with an electrical drier at 50–60°C. Ascospores were observed and measured in KOH. Sections through the peridium were cut anticlinally. All pictures were taken on an Olympus Optiphot-2 microscope. A voucher specimen is deposited in the institutional herbarium of Zoltán Bratek (ZB-3454).

### Soil analysis

One kg of soil was collected by removing the litter and covering vegetation from the spruce forest near Lahti. Soil analyses were performed according to Wedén et al. (2004).

### Sequence analysis

DNA extraction and PCR amplification of ITS-rDNA region was performed according to Kårén et al. (1997) with minor modifications. ITS1 and ITS4 primers (White et al. 1990) were used for PCR and sequencing reactions. For cycle sequencing ABI Prism BigDye™ Terminator Cycle Sequencing Ready Reaction Kit 3.1 (Applied Biosystems) was used. Capillary electrophoresis was carried out on an ABI PRISM 3100 Genetic Analyzer (Applied Biosystems) according to the manufacturer's instructions. The BlastN 2.2.2. program (Altschul et al. 1997) was used to search for published data similar to the monitored sequences in the international database (GenBank-EMBL-DDBJ-PDB). Phylogenetic analysis was performed with MEGA version 3.1 (Kumar et al. 2004). For tree reconstruction Neighbor-Joining analysis was used by default parameters of MEGA programs with one thousand replicates in bootstrap test. *Tuber melanosporum* Vittad. ITS sequence (AF132501) was selected as outgroup based on preliminary phylogenetic analysis. Phylogenetic trees were built using the Neighbor-Joining (NJ) methods (Kumar et al. 2004). The GenBank accession number of the new ITS sequence obtained by this work is FN568055.

## Results

### Ecology

The *T. foetidum* sample was found at a depth of 15 cm in a forest dominated by Norway spruce (*Picea abies*), with scattered birch (*Betula* sp.) and pine (*Pinus sylvestris*). Norway spruce, which comprised 80% of the canopy, averaged 25 m

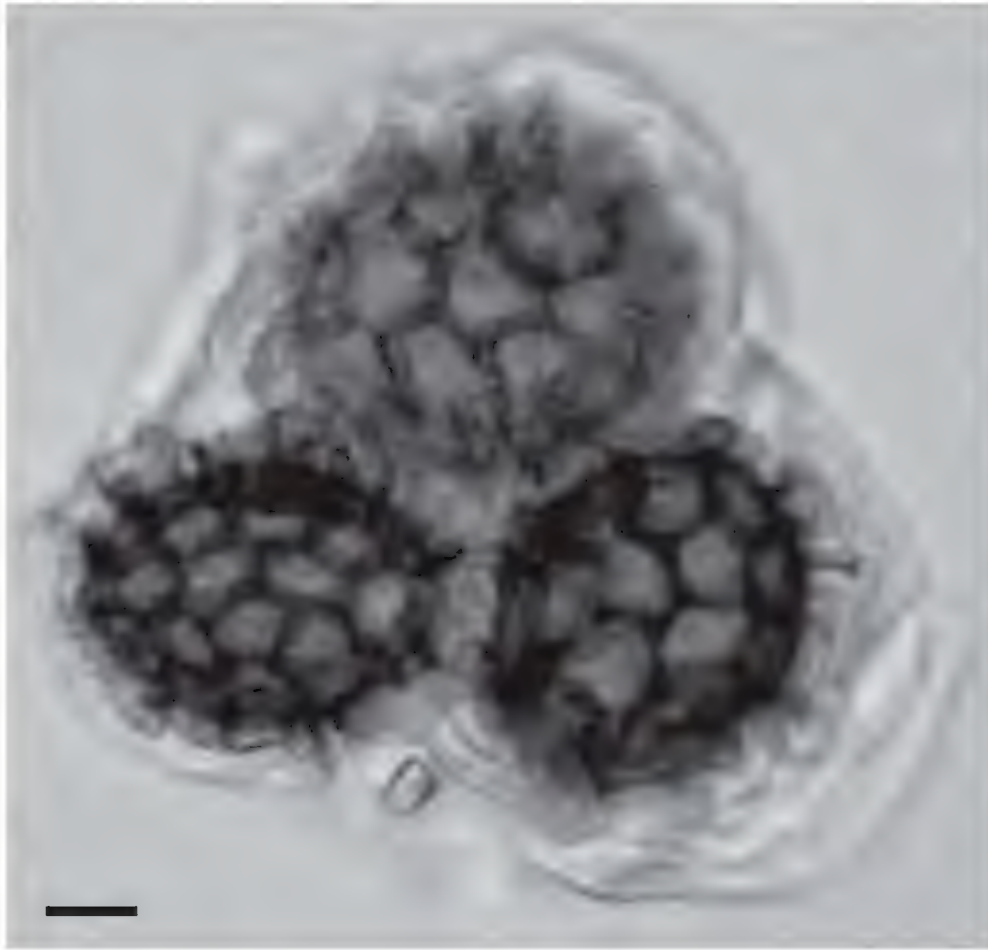


FIGURE 1. Spores of the Finnish *Tuber foetidum* sample.  
Magnification is 100-fold. Scale bar = 10  $\mu\text{m}$ .

in height, 25 cm in diameter, and 40–50 years of age. The soil is cambisol-type silt with a litter layer, lacking stone and organic humus layer. The pH at a ~40 cm measured 6.5; Finnish forest soil pH values generally average 3.5–4.5. The high pH implies that the site may have been used as a farm field or lime fertilizers have been applied earlier.

### Morphology

Ascocarp 9 mm in diam.; surface pale ochraceous brownish, minutely warted to verrucose, not hairy; gleba paler than the surface, rarely marbled; with unpleasant odor. Peridium 330–380  $\mu\text{m}$  thick, pseudoparenchymatous with polygonal or roundish cells 15–19  $\mu\text{m}$  in diam.; cell wall yellow, 0.5–1  $\mu\text{m}$  thick; cystidia lacking. Asci ellipsoid, hyaline, thin-walled, 1–5 spored, lacking a stem; 1-spored asci counting for 20.2%, 2-spored 35.8%, 3-spored 33.9%, 4-spored 9.2% and 5-spored 0.9%. Ascospores ellipsoid, in 1-spored asci: 43.7–36.5  $\times$  38.9–25.5  $\mu\text{m}$ , on average: 40.9  $\times$  31.1  $\mu\text{m}$  (n=10), in 1–2 spored asci 43.3–21.7  $\times$  31.7–21.7  $\mu\text{m}$ , in 4-spored asci 28.7  $\times$  21.3  $\mu\text{m}$ ; spore wall 2–4  $\mu\text{m}$  thick, light golden brown, ornamented with a regular reticulum, formed by mostly hexagonal meshes 3.6–12.2  $\mu\text{m}$  along the spore length and 2.4–8.5  $\mu\text{m}$  across the spore width. A 3-spored ascus is shown in FIGURE 1.

### Sequence analysis

The ITS sequence obtained from the truffle sample covered the entire 560 bp long ITS region; lengths of ITS-1, ITS-2, and 5.8S rRNA gene are 207, 196, and 157 bp, respectively. BLAST searches indicate that the sample sequence matches most closely three identical ITS sequences (FIGURE 2), two from *Tuber foetidum* (AJ557543, AJ557544 in Halász et al. 2005) found in Hungary and one from a *Tuber* sp. (AJ534706) sample found in Estonia (Tedersoo et al. 2003). The 5.8S rRNA gene sequence is identical in our sample and the Hungarian and Estonian *Tuber* materials. Five and two base differences between the Finnish sample and the three above mentioned sequences were found in the ITS-1 and ITS-2 regions, respectively.

### Discussion

In the *Tuber macrosporum* group, *T. foetidum* is known by its stinking odor and verrucose ascocarp surface (Lange 1956, Pegler et al. 1993, Halász et al. 2005). The peridial surface with minute brownish warts, the ellipsoid reticulate spores, and the pseudoparenchymatic peridium of the Finnish specimen correspond to the morphological criteria of *T. foetidum* (RiOUSset et al. 2001, Montecchi & Sarasini 2000).

The N-J tree clustered the Finnish sample into the clade that harbors two Hungarian *T. foetidum* ascocarp samples and the Estonian ectomycorrhizal sample. Inside this clade, the Hungarian and Estonian samples form a well-supported branch with 100% bootstrap support apart from the Finnish sample sequence (FIGURE 2). *Tuber maculatum*, *T. puberulum*, and *T. borchii* (Hungarian samples now deposited in the Zoltán Bratek herbarium; see Halász et al. 2005) clearly belong to a different branch. Despite intraspecific sequence variability of *T. puberulum* sequences from samples originating from different habitats, the *T. puberulum* sequences cluster together with high bootstrap support, just like *T. borchii* sequences. The *T. foetidum* clade (including ZB3454), which shows less variation than the *T. puberulum* clade, is clearly separated from the *T. maculatum*–*T. borchii*–*T. puberulum* groups. For these reasons we classify the Finnish specimen as *T. foetidum*.

ITS sequence differences indicate, however, that the Finnish genotype has begun to evolve apart from the other *T. foetidum* specimens. Further research is needed to explore the origin and status of Finnish *T. foetidum* population. This raises the possibility that *T. foetidum* sequences from other regions might also differ, as suggested by the separation of the two French *T. macrosporum* (FM205664, FM205663) sequences from the other clades. *Tuber foetidum*, which is found in western Europe between 39°N and 62°N (Jeandroz et al. 2008) and has been recorded in the Scandinavian region from Denmark (Lange 1956)

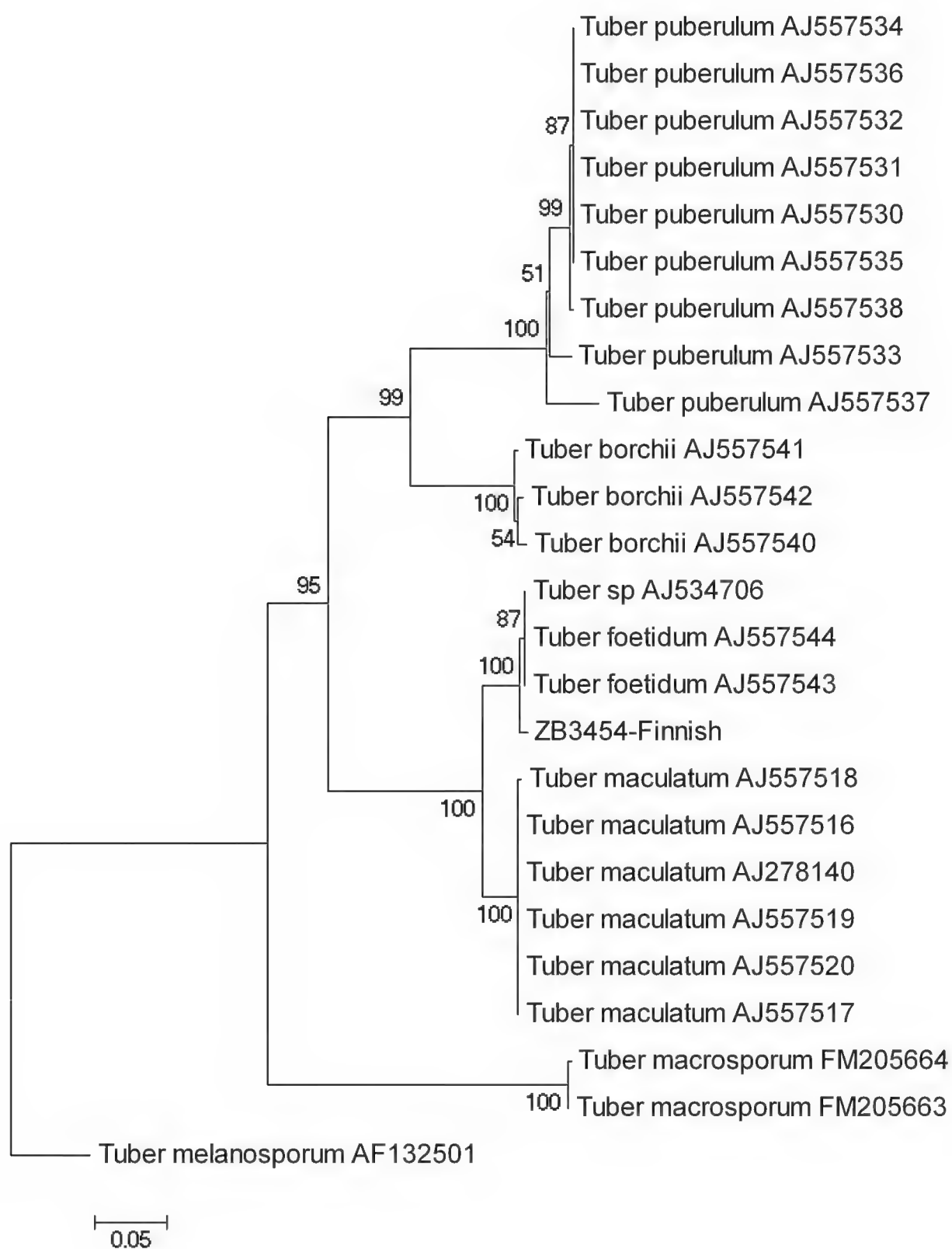


FIGURE 2. Phylogenetic tree based on ITS sequences (ITS-1 and ITS-2). Bootstrap consensus Neighbor-Joining tree based on K-2-p distance matrix (1000 replicates) is shown. Outgroup is *Tuber melanosporum* (AF132501). Scale bar indicates number of nucleotide changes per site.

and Uppland, Sweden (Anderberg & Anderberg 2001) is regarded as a rare species both inside and outside Scandinavia (Lange 1956, Pegler et al. 1993).

*Tuber foetidum* seems to have a broad range of host trees. In southern Europe, it grows in association with fagaceous trees (*Quercus* and *Fagus*) but in the British Isles it has been found in association with *Larix* (Pegler et al. 1993). The truffle was found in deciduous forests with unknown host associations in Denmark (Lange 1956) and under hazel in Sweden (Anderberg & Anderberg 2001). Sequence analyses by Tedersoo et al. (2003) confirm that *T. foetidum* (as “*Tuber* aff. *maculatum*”) formed ectomycorrhizae with birch (*Betula pendula*). Spruce is not a commonly reported host tree for *Tuber* spp. We were unable to trace the ectomycorrhizae, but Norway spruce was the dominant tree in the Finnish forest, we feel that either spruce or Scots pine may serve as hosts of *T. foetidum*.

### Acknowledgements

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**Revisiting the taxonomy of *Daruvedia bacillata***

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**Abstract** — *Daruvedia bacillata*, the type species of the monotypic genus *Daruvedia*, has rarely been collected or reported, but has been placed in many unrelated genera. This paper gives a description of the fungus based on studies of the type specimen, a collection by R.W.G. Dennis, and freshly collected material. The taxon is epitypified and a discussion on its systematic placement is provided.

**Key words** — systematics, *Dothideomycetes*

**Introduction**

We have been carrying out systematic and phylogenetic studies on the *Dothideomycetes* in order to obtain a natural classification system (Zhang et al. 2008a,b,c, 2009a,b,c). This study reports on *Daruvedia* Dennis, an ascomycete genus first proposed for *Sphaeria bacillata* Cooke, a fungus originally collected on decorticated rotten wood by Capron and described in the protologue as a “long-spored sunken *Sphaeria*” (Cooke 1871). This species was later referred to *Acerbia* (Sacc.) Sacc. & P. Syd., *Ceratostomella* Sacc., and *Ophioceras* Sacc.

None of these genera were deemed suitable when Dennis (1988) characterised *Daruvedia* based on a fresh collection from the Hebrides. Dennis (1988) found no ascomata in the type material of *Sphaeria bacillata*, but Cooke's habit sketch and drawings of the ascomata, ascus, and distinctive ascospore on the herbarium packet (FIG. 1) allowed Dennis to identify his new collection as identical with *Sphaeria bacillata* and to propose a new genus for it. Later, Dennis (1989) provided a more detailed account of the taxonomic history and derivation of the word *Daruvedia* but did not assign it to any family or order (Dennis 1988, 1989).

Barr (1994), who studied *Daruvedia* in her survey of North American pyrenomycetes, agreed with Dennis that a separate genus was needed for *Sphaeria bacillata*. Although she did not examine any specimens, Barr (1994) classified *Daruvedia* in the *Pleurotremataceae*, based on her belief that the asci were unitunicate and the genus shared characteristics with other genera included in that family. However, *Pleurotremataceae* sensu Barr is no longer accepted: the 9<sup>th</sup> edition of the Dictionary of the Fungi (Kirk et al. 2001) lists *Daruvedia* as *Dothideales* inc. sed., while the 10<sup>th</sup> Edition (Kirk et al. 2008) lists it as *Dothideomycetes* inc. sed. Eriksson (2006) and Lumbsch & Huhndorf (2007), who retain *Daruvedia* in the *Pleurotremataceae*, classify the family in *Ascomycota* inc. sed. and represented by just two genera, *Pleurotrema* and *Daruvedia*.

We carried out a study using fresh collections, the collection described by Dennis (1988) (designated here as epitype), and the *Sphaeria bacillata* type material to (1) provide a detailed description of this taxon, (2) clarify the taxonomic placement of *Daruvedia bacillata*, and (3) designate an epitype. We also present a preliminary description of an associated coelomycete.

## Materials and methods

Fresh material was collected by Jacques Fournier at different seasons in France. The type specimen of M.C. Cooke and the collection of R.W.G. Dennis were loaned from the Royal Botanic Gardens, Kew (K), UK, for confirmation and more detailed descriptions.

The freshly collected samples were treated following the method used by Hyde et al. (2000) with modification. Dried materials were rehydrated with water first, before checking the morphological characters in water.

Single spore cultures were obtained with the modified method used by Goh (1999).

Total genomic DNA from specimens was extracted directly from ascomata by using Forensic Kits following the instructions. The genomic DNA from cultures was extracted following a protocol as outlined by Cai et al. (2005, 2006). Polymerase chain reaction (PCR) amplification products were obtained with the two pairs of primers, ITS4 and ITS5 (White et al. 1990) and LROR and LR5 for partial rDNA LSU (Vilgalys & Hester 1990).



FIG.1. Cooke's drawings of *Sphaeria bacillata* from the holotype (K).

## Results

After examining the specimens, including the drawings of Cooke on the herbarium packet (FIG. 1) and those of Dennis (1988), we concur with Dennis that *Daruvedia* should be maintained as a distinct genus for *Sphaeria bacillata*.

We found that Cooke's type material lacked ascomata as Dennis (1988) had mentioned, but Dennis's material is still in good condition. Here we provide a detailed description based on Cooke's drawing, Dennis's specimen and drawing, and our recent collections from France and designate Dennis's specimen as an epitype.

***Daruvedia bacillata*** (Cooke) Dennis, *Belarra* 2(4): 25, 1988.

FIGS. 2–4

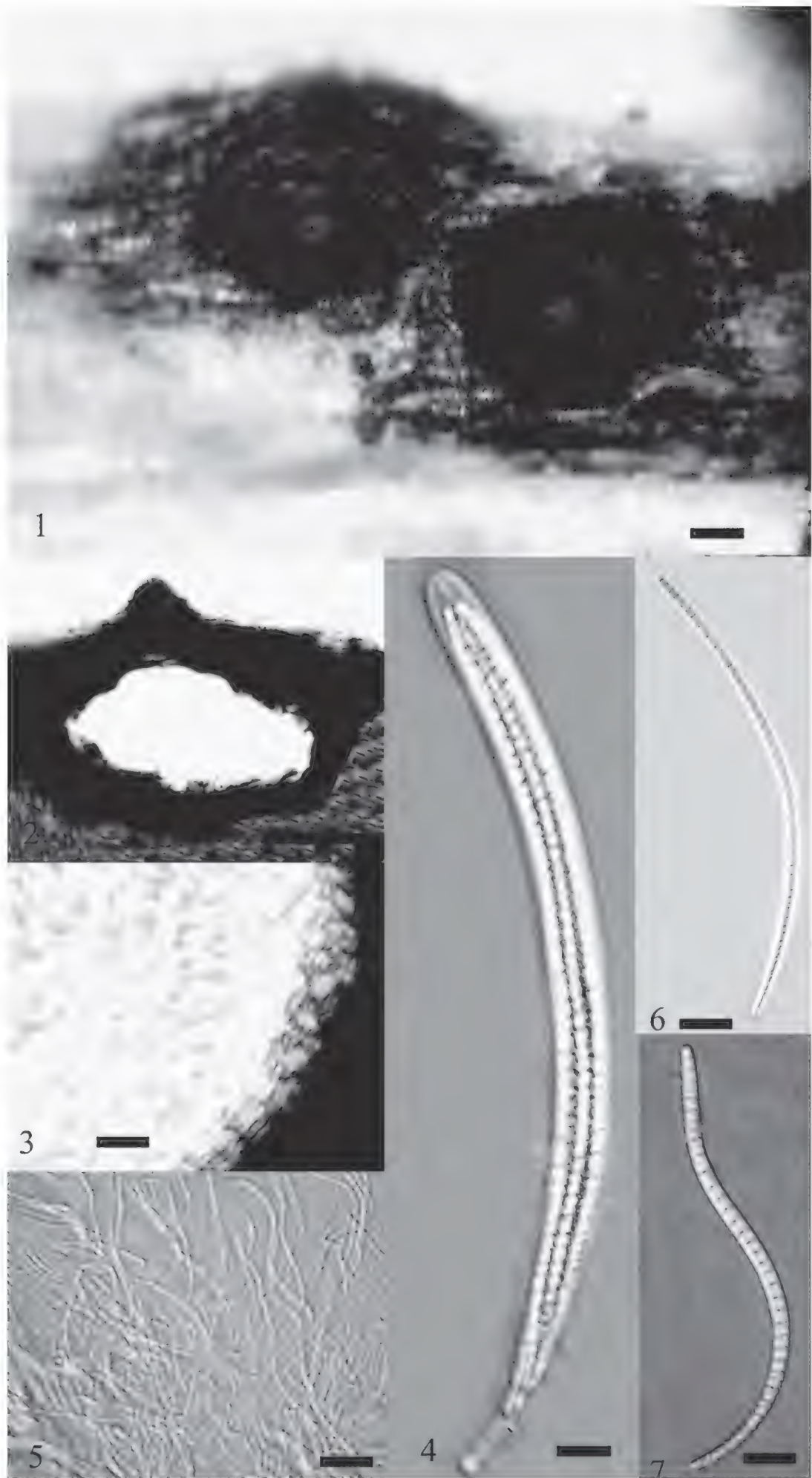
≡ *Sphaeria bacillata* Cooke, *Handbook of British Fungi* 2: 879, 1871.

≡ *Ophioceras bacillata* (Cooke) Sacc., *Sylloge Fungorum* 2: 360, 1883.

≡ *Ceratostomella bacillata* (Cooke) Cooke, *Grevillea* 17: 50, 1889.

≡ *Acerbia bacillata* (Cooke) Berl., *Icones Fungorum* 2: 142, 1899.





= *Rhaphidophora macrocarpa* Sacc., Nuovo Giornale Botánico Italiano 7: 306, 1875.

= *Ophioceras macrocarpum* (Sacc.) Sacc., Sylloge Fungorum 2: 359, 1883.

Ascomata scattered, rarely gregarious, erumpent through bark or wood, immersed to nearly superficial with base remaining immersed in the host tissue (FIGS. 2.1, 3.1), depressed spherical, subglobose, broadly or narrowly conical, black, roughened, 500–1000 µm high, 350–800 µm diam.; apex obtuse, pointed, discoid-flattened, up to 250 µm high, 200 µm broad (FIGS. 2.2, 3.2), sometimes hardly protruding in case of small fully immersed ascomata, and then surrounded by a black clypeus-like disc. The discs often bear tufts of brown hairs seated on an easily removed cushion-like structure; this material (a setose acervular coelomycete) was found to belong to a different taxon (see DISCUSSION) and does not represent an anamorphic state of *D. capillata* (FIGS. 3.1, 4.1). Peridium 30–40 µm thick for immersed parts, 60–100 µm thick above, two-layered; outer layer nearly homogenous, of very thick-walled cells with small lumina, inner layer textura prismatica, about 25 µm thick, of flattened cells 7–12 × 2.2–5 µm with unevenly pigmented walls, giving the appearance of alternating dark and pale columns oriented perpendicular to the surface (FIGS. 2.3, 3.3). Hamathecium of dense, very long pseudoparaphyses, 1–1.5 µm broad, sparse (FIGS. 2.5, 3.4). Asci 240–270 × 15–17 µm, 8-spored, bitunicate, but not fissitunicate, cylindrical to fusiform, short stipitate, with a narrow ocular chamber and a small inconspicuous apical apparatus (FIGS. 2.4, 3.5). Ascospores 180–200 × 4–5 µm, filiform, apex obtusely rounded without evident mucilage, base slightly tapered with inconspicuous mucilaginous material on some spores, yellowish, lying parallel in the ascus, filled with guttules, obscurely 30–40-septate, slightly constricted at septa at full maturity, smooth-walled (FIGS. 2.6–7, 3.6).

SPECIMENS EXAMINED: ENGLAND. SURREY: Shere, on dead stick, probably *Hedera*, leg. Capron 1567, M.C. Cooke (K, **holotype** of *Sphaeria bacillata*). SCOTLAND. ISLE OF ISLAY: Bridgend, Islay House, on dead stem of *Hedera helix*, old garden wall, 24 Jun. 1987, R.W.G. Dennis (K, **epitype** of *Sphaeria bacillata* **designated** here). FRANCE. Ariège, Rimont, Las Muros, on *Lonicera nigra*, 9 Jun. 1996, JF 96083; same locality, on *Acer campestre*, 17 Jun. 1996, JF 96086; same locality, on *Cornus sanguinea*, 2 Mar. 1997, JF 97057; Rimont, Le Baup, on *Populus tremula*, 14 Apr. 2005, JF 05050; same locality, on *Frangula alnus*, 14 Apr. 2005, JF 05051; Rimont, Peyrau on *Acer campestre*, 5 Oct. 2005, JF 05126; Rimont, Las Muros, on twigs of *Hedera helix*, 7 December 2005, leg. J. Fournier, det. Paul Leroy, JF 05159; same locality, on twigs of *Hedera helix*, 14 Mar. 2007, JF07026; same locality, on dead twigs of *Clematis vitalba*, 470m, 19 Jun. 2008, JF 08144; same locality, on dead decorticated twig of *Hedera helix*, 470 m, 1 Jul. 2008, JF 08155.

FIG. 2 (at left). Dennis's collection of *Daruvedia bacillata* from Scotland (K). 1. Ascomata on substrate. 2. Section of ascoma. 3. Peridium. 4. Ascus. 5. Pseudoparaphyses. 6, 7. Ascospore. Scale bars: 1, 2 = 100 µm, 3–7 = 10 µm.

## Discussion

*Daruvedia bacillata* is uncommon but has been found on various hosts in diverse families: *Acer campestre* (Aceraceae), *Clematis vitalba* (Ranunculaceae), *Cornus sanguinea* (Cornaceae), *Frangula alnus* (Rhamnaceae), *Hedera helix* (Araliaceae), *Lonicera nigra* (Caprifoliaceae), and *Populus tremula* (Salicaceae), mainly on decorticated wood. Its occurrence is perennial. The ascoma shape and degree of immersion in the substrate are highly variable. The striking wig-like conidial hairy structure at the apex of the ascomata, which is fragile, easily removed, and often absent when fully mature, appears to be an associated fungus.

When Cooke (1871) first described this fungus, he provided a drawing of the ascomata, one ascus, and one ascospore. Later, Dennis (1988) described a new genus *Daruvedia* for this fungus, but did not assign the genus to any family or order. As *Daruvedia capillata* has bitunicate asci, it does not belong in unitunicate *Pleurotremataceae* in the broad sense of Barr (1994). Because mature fruitbodies of *D. capillata* remain embedded in sterile tissues, the genus does not belong in *Dothideales* (Kirk et al. 2001), which is characterised by the lack of hymenium when mature. Based on our results we agree with Kirk et al. (2008), who placed *Daruvedia* in the *Dothideomycetes* in the *Pleosporales*; further molecular work is needed to place the fungus in a suitable family.

We tried to isolate *Daruvedia bacillata* from single spores and conidia from the setose acervular coelomycete but failed. However, we were able to obtain pure cultures from the ascomatal spore mass. We sequenced the fungus from the cultures and were surprised when the sequence data blasted closest to *Exophiala pisciphila* McGinnis & Ajello (ITS: AF050272; LSU: DQ823101). Extraction of DNA directly from the ascomata produced the same result. As *Exophiala* spp. have teleomorphs in *Capronia* (a genus with short, fusiform, 1–2-celled ascospores), it was obvious that our isolated fungus and sequences do not derive from *Daruvedia capillata*, which has long filiform ascospores. A prolonged dry period in Pyrenees prevented our collecting more material for further study.

No anamorph has been linked to *Daruvedia bacillata*. In this study we found one associated anamorphic taxon that may represent a hyperparasite on *Daruvedia bacillata*. This unidentified fungus has acervuli comprising numerous cylindrical, long and narrow ( $150\text{--}200 \times 4\text{--}5 \mu\text{m}$ ), thick-walled, nonseptate, brown hairs with paler obtuse ends that are often constricted just beneath the apex (FIG. 4.1) and which arise from a basal brown pseudoparenchymatous tissue with the hairs aggregating into stellate tufts (FIG. 4.2–3). Other characters include  $< 2 \mu\text{m}$  diam. conidiophores that form at the base of the hairs and are composed of palisade or dense ramified, hyaline bunches (FIG. 4.4–5);, percurrent, denticulate,  $< 2 \mu\text{m}$  diam. conidiogenous cells (FIG. 4.4–5), and



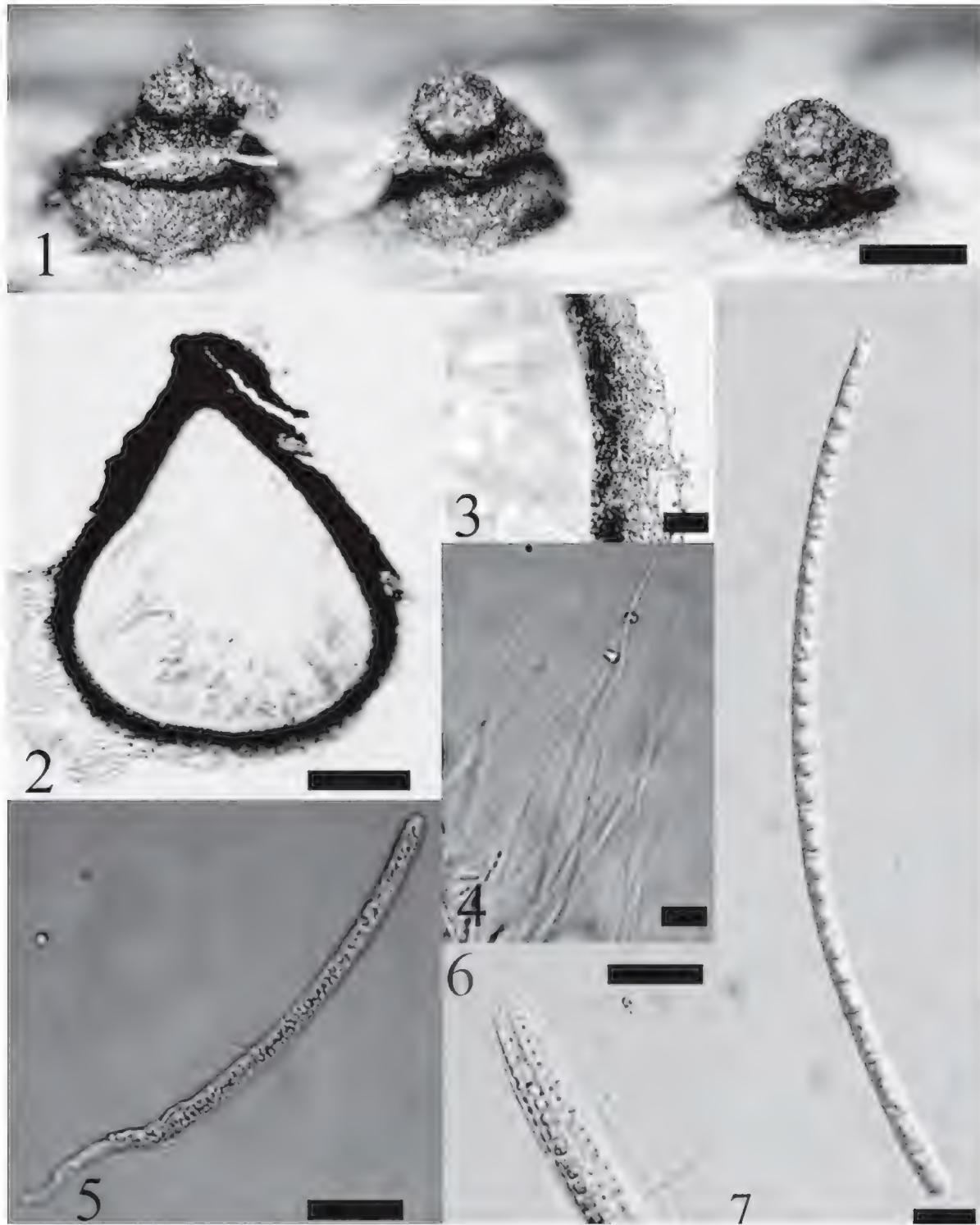


FIG. 3. Collections of *Daruvedia bacillata* from France (JF05159). 1. Mature ascoma on host surface. 2. Section of ascoma. 3. Peridium. 4. Pseudoparaphyses. 5. Ascus with ascospores. 6. Apical portion of an ascus showing ocular chamber. 7. Ascospore. Scale bars: 1 = 300  $\mu\text{m}$ , 2 = 200  $\mu\text{m}$ , 3 = 10  $\mu\text{m}$ , 4 = 10  $\mu\text{m}$ , 5 = 50  $\mu\text{m}$ , 6 = 20  $\mu\text{m}$ , 7 = 10  $\mu\text{m}$ .

ovoid, hyaline, conidia with narrow hila and measuring  $3.5\text{--}4 \times 1.8 \mu\text{m}$  (FIG. 4.5–6). This structure is only usually present on young erumpent ascomata and has always been dislodged from older mature ascomata. Further research is needed to establish the nature of the association between *D. capillata* and the unknown coelomycete.

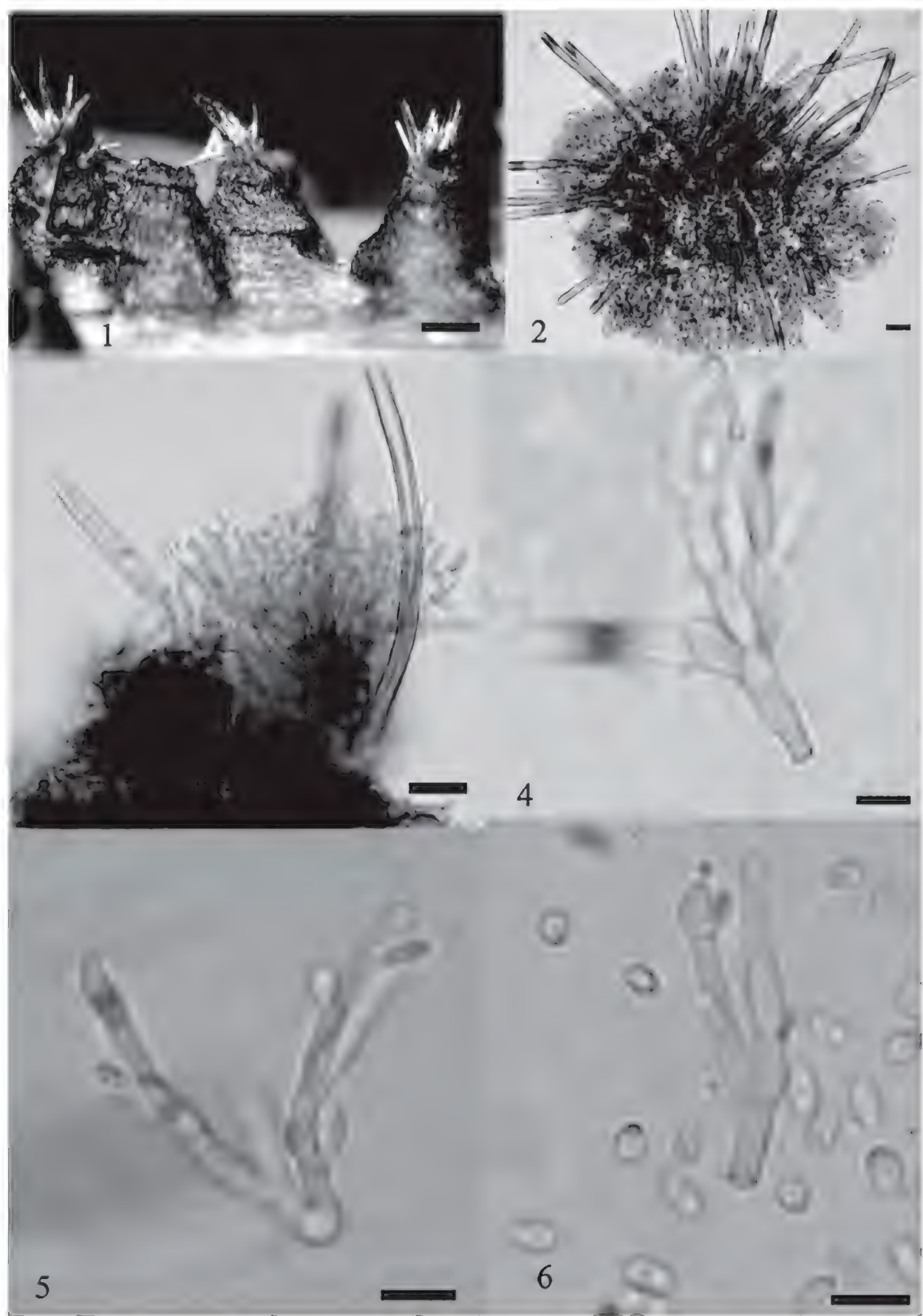


FIG. 4. Coelomycetous fungus associated with *Daruvedia bacillata*. 1. Immature *Daruvedia* ascomata with associated unknown conidiomata on natural substrate. 2, 3. Acervulus. 4–6. Conidiogenous cells and conidia from natural substrate. Scale bars: 1 = 200  $\mu\text{m}$ , 2–6 = 10  $\mu\text{m}$ .



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Hu Hongli thanks the University of Hong Kong for the award of a postgraduate studentship. Helen Leung is thanked for her kind laboratory assistance. K.D. Hyde thanks BRT grant number R253012 for the award of a scholarship to study *Dothideomycetes*. Wen-Ying Zhuang, De-Qun Zhou, Eric McKenzie are thanked for the pre-submission reviews of our manuscript.

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## MYCOTAXON

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**Observations on the *Bolbitiaceae* 31.  
*Conocybe volviradicata* sp. nov.**ROY WATLING<sup>1</sup>, MUSTAFA İŞILOĞLU<sup>2</sup> & HAYRÜNİSA BAŞ SERMENLİ<sup>2\*</sup><sup>1</sup> *caledonianmyc@blueyonder.co.uk**Caledonian Mycological Enterprises, Edinburgh, EH4 3HU, Scotland*<sup>2</sup> *isiloglu48@gmail.com* & *\*hayba2000@gmail.com**Muğla University, Faculty of Science and Arts, Biology Department  
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**Abstract** — A new species of *Conocybe* from southwest Turkey, with the unique combination of a volva and long radicating stipe-base, is described as new to science; it is placed in *Conocybe* section *Singerella*.

**Key words** — new taxa, *Conocybe corneri*, *Conocybe antipus*

**Introduction**

Other than *Conocybe peronata* Kühner & Watling, now assigned to *Pholiotina*, no peronate or volvate species were treated in the classical studies of the genus by Atkinson (1918; as *Galerula*) and Kühner (1935). Watling (1979) was the first to describe two species from South East Asia with a distinct volva — *C. corneri* Watling and *C. vaginata* Watling — and to transfer *Galerula locellina* Murrill from Florida, North America, to *Conocybe*, noting that it also possessed a volvate stipe-base. Over the intervening years, a clutch of taxa have been recognized with this character, with Horak & Hausknecht (2002) eventually providing a key to nine species. Since then Hausknecht & Krisai-Greilhuber (2009) have added a tenth species, *C. reinwaldii*. Whilst documenting the mycota of southwest Turkey, we discovered a new member of this group that differed from all others by possessing a radicating stipe-base. This new taxon, formally described herein, is the twelfth representative of *Conocybe* in the Turkish macromycota (Solak et al. 2007).

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## Taxonomy

***Conocybe volviradicata*** Watling, Işiloğlu & Baş Sermenli, **sp. nov.**

FIGS 1–4

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*Pileus 15 mm, e convexo vel campanulato rariore expansus clare cinnamomeus vel ferrugineo-mellinus siccitate bubalinus vel flavo-cremeus glabrus ad marginem striatus tenuis. Lamellae fere liberae aggregatae mellino-luteolobrunneae. Stipe 30 × 3 mm, tenax cremeus conspicue pruinoso-striatus ad basim leviter incrassatus volvatus et radicans (< 20 mm longus). Caro tenuis. Sporae in cumulo ochraceo-brunneae vel cinnamomeae. Sporae hexagoneae poro germinativo 8–10 × 6–7 µm. Cystidia aciei lamellarum lecythiformia 22–25 × 4–7 µm. Cystidia stipitis 1) lecythiformia 25–35 × 2–6 µm; 2) ellipsoidea vel clavata 18–21 × 6–8 µm; et 3) utriformia vel lageniformia 25–30 × 6–9 µm. Fibuligeris nullus. Habitatio in fimo putrido. Turkey; Muğla, Göktepe. Typus H. Baş 12 in E.*

TYPE: Turkey; Muğla, Göktepe village, 11 September 2004, Işiloğlu 7700, H. Bas 12.

HOLOTYPE: in E.

ETYMOLOGY: The epithet *volviradicata* refers to the volvate, rooting stipe.

PILEUS 15 mm conical to campanulate (FIG. 1), deep cinnamon to sienna, drying buff to yellowish cream, smooth, margin striate, silky and very thin. GILLS almost free, crowded, pale ochraceous. STIPE 30 × 3 mm, tough, cream-colour, distinctly striate to beginning of the volva, volvate, rooting base < 20 mm long. FLESH thin, < 1 mm thick in cap-center. TASTE AND SMELL not recorded. SPORE PRINT ochraceous-brown to cinnamon. SPORES hexagonal (FIG. 2) 8–10 × 6–7 µm, sienna, thick walled, with a distinct germ-pore. CHEILOCYSTIDIA lecythiform (Fig. 3) 22–25 × 4–7 µm. CAULOCYSTIDIA mixed of 3 different types: 1) lecythiform, 25–35 × 3–6 µm; 2) ellipsoid to clavate, 18–21 × 6–8 µm; 3) nettle hair-shaped to lageniform, 25–30 × 6–9 µm (FIG. 4). CLAMP CONNECTIONS not seen.

HABITAT. On manured soil bordering a vegetable garden.

## Discussion

*Conocybe volviradicata* is very easily recognized in the field by its distinct membranous volva and long, radicating stipe-base. The presence of lecythiform cheilocystidia combined with the field characters places this new species firmly in section *Singerella* Watling, and the presence of lecythiform caulocystidia places it in a slightly modified series *Corneri* Hauskn. & Krisai, as outlined by Hausknecht & Krisai-Greilhuber (2006). The stipitipellis in series *Corneri* consists of capilliform, ellipsoid, and sphaerical to lageniform elements; only in one species are these elements intermixed with lecythiform cells.

The volva in *C. volviradicata* is striate on the upper surface, and although it has been impossible to track the development to the degree followed by the senior author for *C. corneri* (Watling 1979), a striate volva characterizes both species. There are other parallels. *Conocybe corneri* is coprophilous, with the primordia

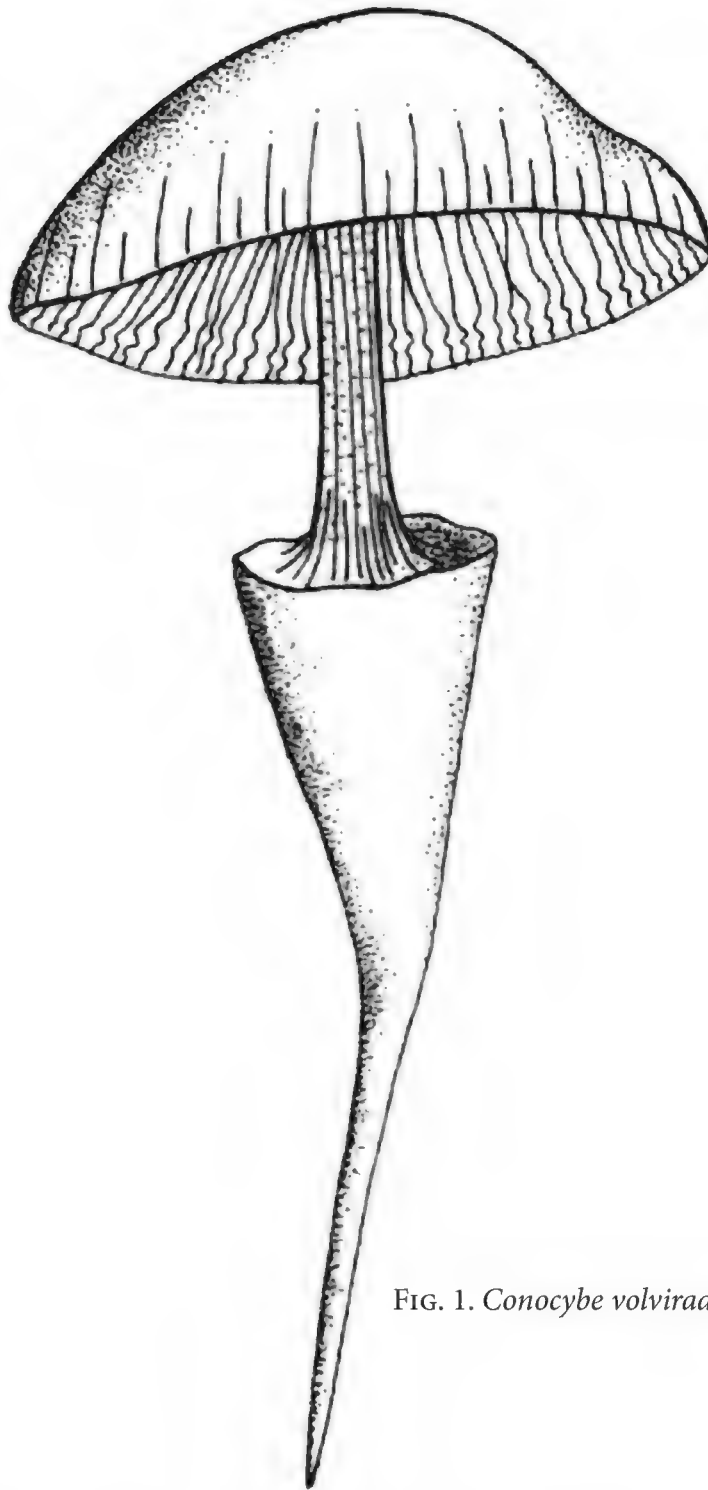
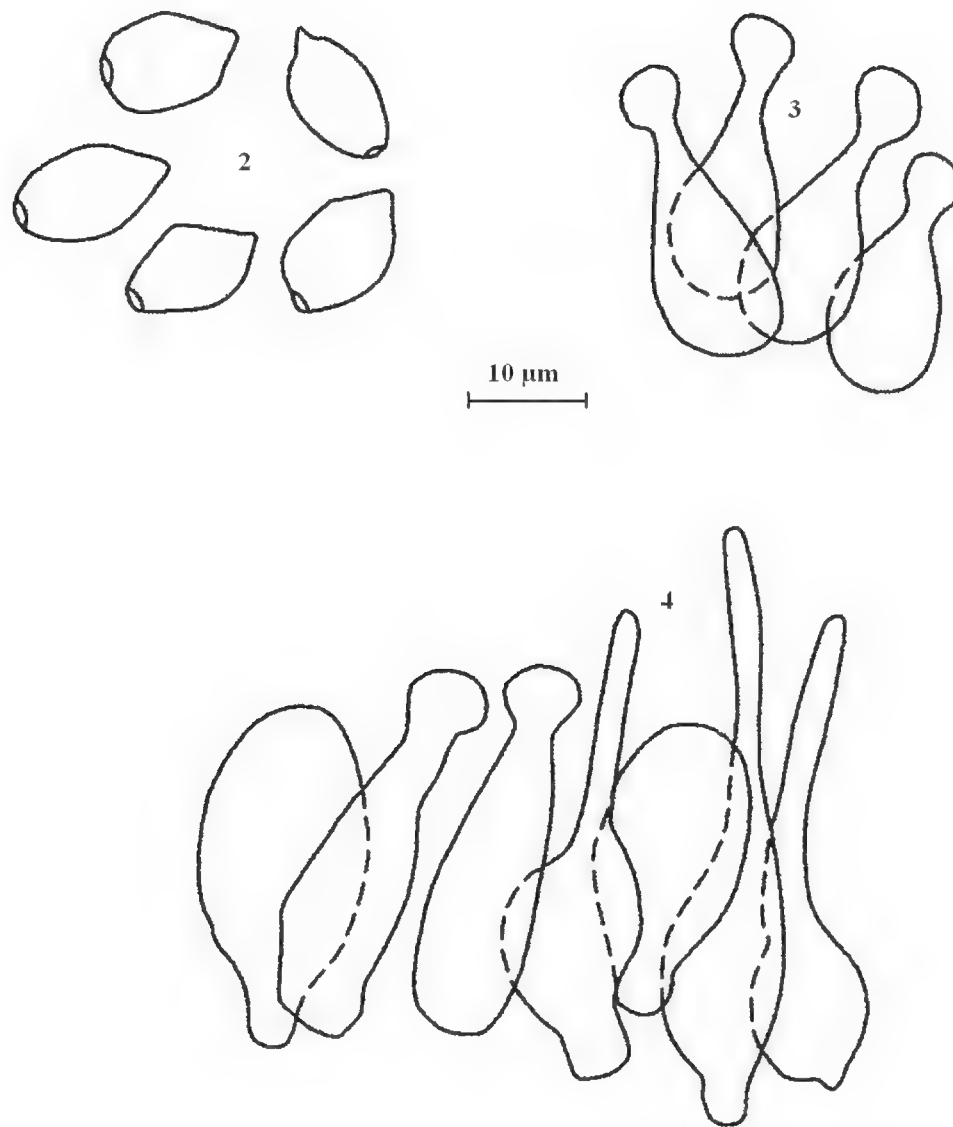


FIG. 1. *Conocybe volviradicata*. Habit.

developing below the surface of the dung, whereas in *C. volviradicata* the stipe was found buried in manured soil.

The long rooting base of *C. volviradicata* resembles that found in *C. antipus* (Lasch) Fayod known from Europe and North America, *C. humicola* (Thiers) Hauskn. et al. ( $\equiv$  *C. antipus* var. *humicola* Thiers) from North America, and the European *C. fiorii* (D. Sacc.) Watling, *C. leporina* (Velen.) Singer, and *C. alboradicans* Arnolds—all assignable to *Conocybe* series *Antipus* (Hausknecht & Krisai-Greilhuber 2006). Although none of these species ever develop a volva





FIGS. 2–4. *Conocybe volviradicata*. 2. Basidiospores. 3. Cheilocystidia. 4. Caulocystidia.

immediately above or at the base of the rooting stipe, *C. volviradicata* resembles *C. antipus* in producing basidiospores that are hexagonal in face view.

Hausknecht (1996, 2009), who treated European *Conocybe* species with rooting or deeply inserted stipe-bases including the species indicated above, recognized eight additional species in his key but did not depict any possessing the slightest volvate development. The recently described *C. reinwaldii* from Europe (Hausknecht & Krisai-Greilhuber 2009) and *C. radicata* Singer, an extra-European radicate taxon with minutely ornamented basidiospores, are placed in *Conocybe* section *Ochromarasmius* subsection *Pseudocystidiatae*. The spores of *C. volviradicata*, however, are smooth. *Conocybe reinwaldii* differs significantly in the lack of a volva.

*Conocybe radicata* from South America possesses lecythiform pleurocystidia, but no such structures are found in *C. volviradicata*. *Conocybe radicata* is also lignicolous, whereas the Turkish material is found in manured garden soil.

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## MYCOTAXON

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***Postia stellifera* sp. nov.,  
a stipitate and terrestrial polypore from Malaysia**

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**Abstract** — *Postia stellifera* sp. nov. from Malaysia is described. This fungus is characterized by the distinctly stipitate basidiomata, terrestrial habit, and verrucose chlamydospores both in the context and in culture. Its macromorphology resembles that of *Albatrellus*, but phylogenetic analysis based on LSU places it within a clade comprising *Postia*, *Amylocystis*, and *Jahnoporus* where all species have a white and fleshy to soft corky context and monomitic hyphal system with clamped generative hyphae. Most sequences showing high homology with *P. stellifera* represent brown rot polypores.

**Key words** — *Fomitopsidaceae*, *Oligoporus*, phylogeny, *Polyporaceae*, taxonomy

## Introduction

*Postia* Fr. (*Fomitopsidaceae*, *Polyporales*) is typified by *Polyporus lacteus* Fr. [lectotypified by Donk (1960); = *Postia tephroleuca* (Fr.) Jülich]. The genus is characterized by resupinate to sessile basidiomata with a fleshy context in fresh condition, a monomitic hyphal system with clamped generative hyphae, and causing a brown rot. A few species such as *P. ceriflua* (Berk. & M.A. Curtis) Jülich, *P. folliculocystidiata* (Kotl. & Vampola) Niemelä & Vampola, and *P. subundosa* Y.L. Wei & Y.C. Dai occasionally produce substipitate or pendent

basidiomata (Ryvarden & Gilbertson 1994, Wei & Dai 2006), but so far, species with terrestrial and distinctly stipitate basidiomata are unknown in the genus.

*Oligoporus* Bref. has been used for the same group of fungi (Ryvarden 1991), but *Postia* was published prior to *Oligoporus* and has been widely accepted (Buchanan & Ryvarden 2000, Dai et al. 2004, Dai et al. 2007, Niemelä et al. 2001, Rajchenberg 2006).

*Tyromyces* P. Karst. (type: *Tyromyces chioneus* (Fr.) P. Karst.) is morphologically similar to *Postia*, and many species now accommodated in *Postia* were once placed in *Tyromyces* (Lowe 1975, Ryvarden 1978). *Tyromyces* has been restricted to species producing a white rot, however. Phylogenetic studies also suggest that *Tyromyces* is phylogenetically distinct from *Postia* (Binder et al. 2005, Yao et al. 1999).

During field trips in Peninsular Malaysia in 2002 and 2007, we collected a polypore with distinctly stipitate and terrestrial basidiomata, a white and fleshy context, oblong ellipsoid basidiospores, and verrucose chlamydospores in the context. Its mycelium in pure culture did not react with 1-naphthol, suggesting a lack of laccase and, consequently, that it is not a white rot fungus (Käärik 1965).

Within the genera of polypores (Ryvarden 1991), the micro-morphological and physiological features of this species would point toward *Postia*. However, the terrestrial and stipitate habit together with the verrucose chlamydospores deviates from *Postia* as currently circumscribed.

In this study, we examined the phylogenetic position of the present fungus in relation to several *Postia* spp. and related polypores. After detailed morphological examinations and other characteristics, we describe it as a new species.

## Materials and methods

### Sequencing and phylogenetic analysis

Five fungal isolates including *Postia* spp. (TABLE 1) were grown and harvested according to Ota & Hattori (2008). DNA was extracted using a DNeasy Plant Mini Kit (Qiagen, Valencia, CA, USA). Nuclear ribosomal LSU sequences were generated following the methods of Ota & Hattori (2008) or Sotome et al. (2008). DNA sequences were determined using a BigDye Terminator 3.1 Cycle Sequencing Kit (Applied Biosystems, Foster City, CA, USA) with the ABI 3100 DNA sequencer (Applied Biosystems). Sequences were edited with Vector NTI advance 9.0 (InforMax, Frederick, MD, USA) then submitted to GenBank (accession numbers AB569119-569123, Table 1). Twelve additional nrLSU sequences were retrieved from GenBank. *Lentinus tigrinus* (Bull.) Fr. and *Polyporus squamosus* (Huds.) Fr. were used as outgroups because they belong to *Polyporaceae* but are outside of the family *Fomitopsidaceae* that accommodates *Postia* species and their allies. The sequences were aligned using Clustal X (Thompson et al. 1997). The alignment of the nrLSU regions was deposited in TreeBase (accession



TABLE 1. List .of species, strains, and voucher specimens newly sequenced in this study and GenBank accession numbers for the LSU sequences.

SPECIES	STRAIN NO.	VOUCHER NO.	ORIGIN	ACCESSION NO.
<i>Postia caesia</i>	WD-1974	F-18596	Japan, Kochi	AB569119
<i>P. caesia</i>	WD-1976	F-18605	Japan, Kochi	AB569120
<i>P. japonica</i>	WD-2103	F-19345	Japan, Kyoto	AB569121
<i>P. japonica</i>	WD-2338	IFP Dai 8046	Japan, Ibaraki	AB569122
<i>P. stellifera</i>	PEN49	F-20668	Malaysia, Penang	AB569123

S10658). The data set was analyzed in PAUP\* 4.0b10 (Swofford 2003). Maximum parsimony analysis was performed for the dataset with the heuristic search option with 100 random addition sequences and tree bisection and reconstruction (TBR) as the branch-swapping algorithm. All gaps were treated as missing data. The robustness of individual branches was estimated based on 1000 bootstrap replications.

Morphological studies

Macroscopic characteristics were described based on fresh and dried specimens. Microscopic characteristics based on dried specimens were determined by examining free-hand sections mounted in Melzer’s reagent or in 5% (w/v) KOH solution. A non-dextrinoid and non-amyloid reaction was described as IKI–. The following abbreviations are used in the text: L, mean spore length; W, mean width; r, the ratio of length/width of a basidiospore; R, mean of r. The term (n = x/y) means x measurements of basidiospores from y specimens. The examined specimens were deposited in TFM or KEP.

Cultural characteristics were studied on potato dextrose agar plates at 25°C and described according to Nobles (1965) and Stalpers (1978). Presence of extracellular oxidase was tested with 1-naphthol ethanol solution and tyrosine ethanol suspension (Käärik 1965). The examined culture was deposited in the culture bank of Forestry and Forest Products Research Institute (FFPRI), Tsukuba, Japan.

Results

Phylogenetic analysis

A preliminary search using the blast option showed homology with several brown rot polypores. The phylogenetic affinities of the present fungus were estimated using 20 LSU sequences, with an aligned length of 751 base pairs. Fifty positions were variable but uninformative and 86 positions parsimony were informative. Parsimony analysis of the nrLSU data set yielded two most parsimonious trees, 269 steps in length (CI = 0.58, RI = 0.68, RC = 0.40) (Fig. 1).

The present fungus was placed within a weakly supported clade that includes the species *Postia caesia* (Schrad.) P. Karst., *P. guttulata* (Peck) Jülich, *P. japonica* Y.C. Dai & T. Hatt. and *P. rennyi* (Berk. & Broome) Rajchenb. This clade is included in a larger one (*Postia* s.l. clade) that includes *Amylocystis*

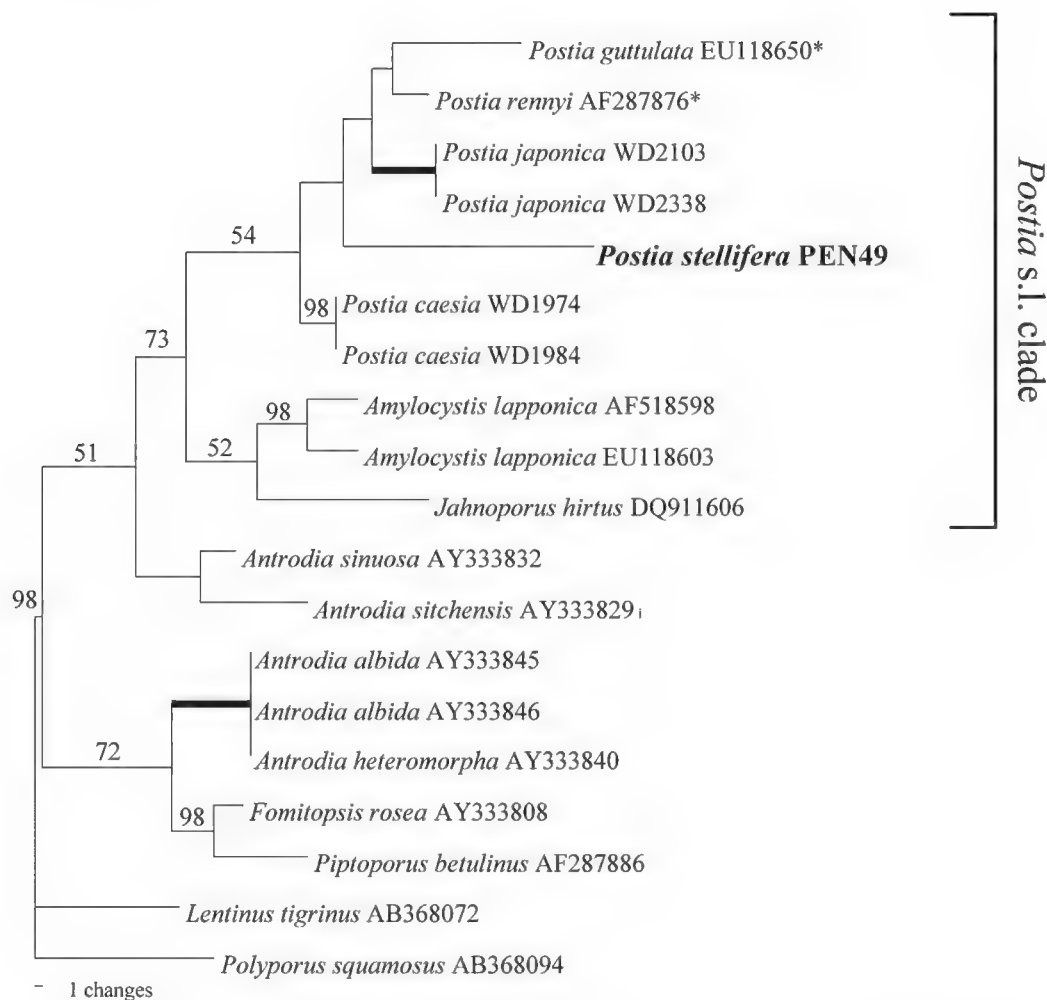


FIG. 1. One of the two most parsimonious trees obtained from heuristic searches based on LSU sequence dataset of *Postia stellifera* and its related species. Bootstrap support values above 50% are indicated at the nodes. Taxa marked with \* were originally submitted to GenBank as *Oligoporus*.

*lapponica* (Romell) Bondartsev & Singer ex Singer, and *Jahnoporus hirtus* (Cooke) Nuss. The cultural characteristics of *J. hirtus* are still not fully known, but other members of this clade are brown rot polypores with a monomitic hyphal system.

Description

*Postia stellifera* T. Hatt. & Sotome, sp. nov.

FIGS 2, 3

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*Basidiocarpia annua, stipitata, terrestria. Pilei circulares, subtomentosi, brunneoli. Contextus carnosus, albus. Facies pororum alba, pori angulares, (1–)2–3 per mm. Stipes centrales, albi. Systema hypharum monomiticum, hyphae generativae hyalinae, fibulatae, hyphae in contextu inflatae. Basidiosporae oblongae, hyalinae, haud dextrinoideae, 4.5–5.5 × 1.8–2.3 μm. Chlamydosporae verrucosae, hyalinae vel luteolae, 7.5–12.5 × 6.8–10.8 μm.*

HOLOTYPE: **Malaysia.** Penang, Gertak Saggul, ad terram in silva, 26.XII.2002, leg. T. Hattori & S. Baharuddin (TFM F-20668).

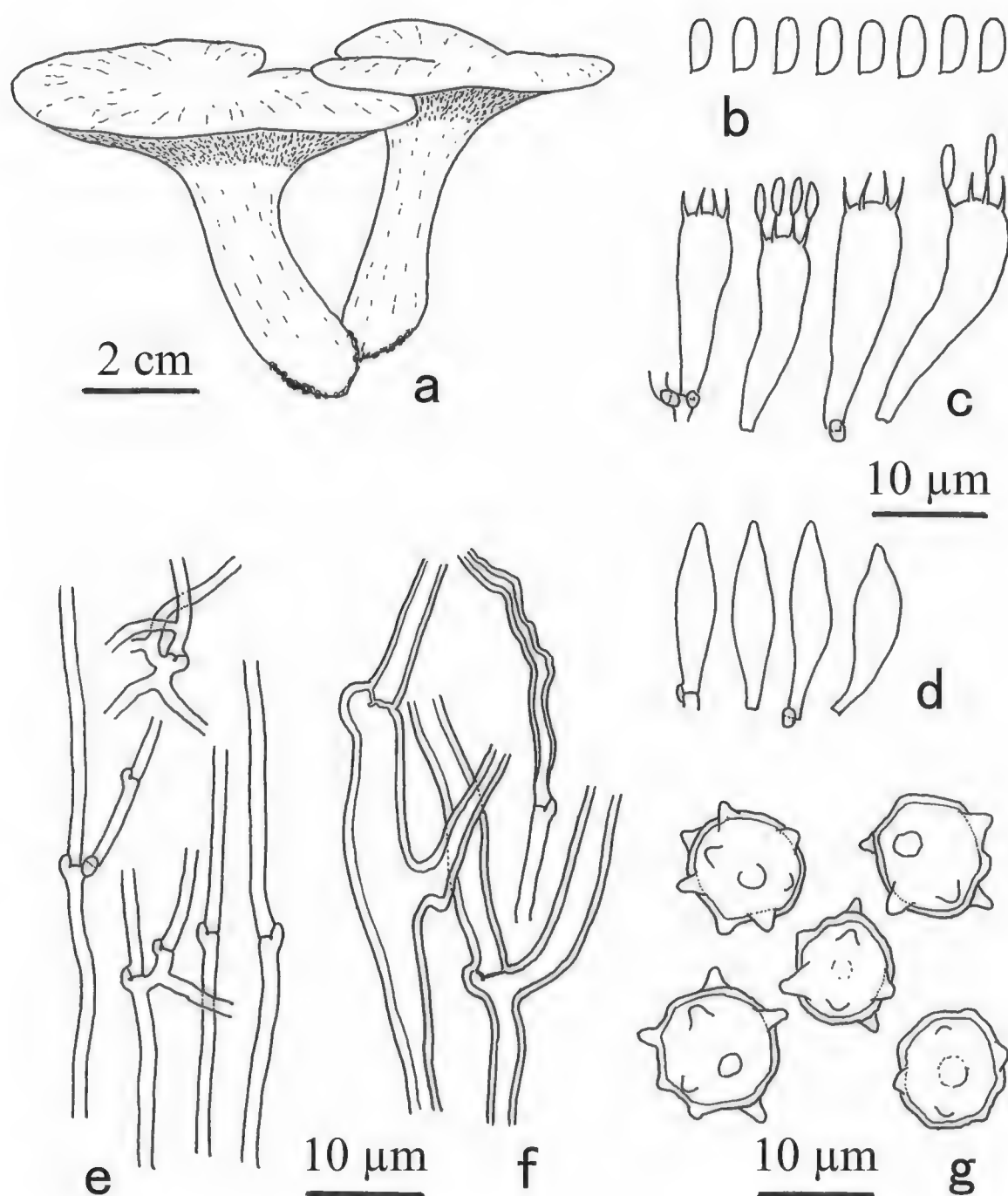


FIG. 2. Structures of *Postia stellifera* (from holotype).  
—a: Basidiocarps. —b: Basidiospores. —c: Basidia. —d: Cystidioles. —e: Chlamydospores from context. —f: Generative hyphae from trama. —g: Generative hyphae from context.

ETYMOLOGY: Latin, *stellifera* = with stars, referring to the star-shaped chlamydospores seen both in the context of the basidiomata and in the culture.

Basidiomata annual, centrally stipitate, terrestrial. Pilei circular, applanate to convex, pileus surface subtomentose to pubescent, azonate, light brown to light grayish brown, whitish near the margin, pileus margin thin and acute, entire, up to 7 cm in diam. Pore surface white to cream in fresh condition drying

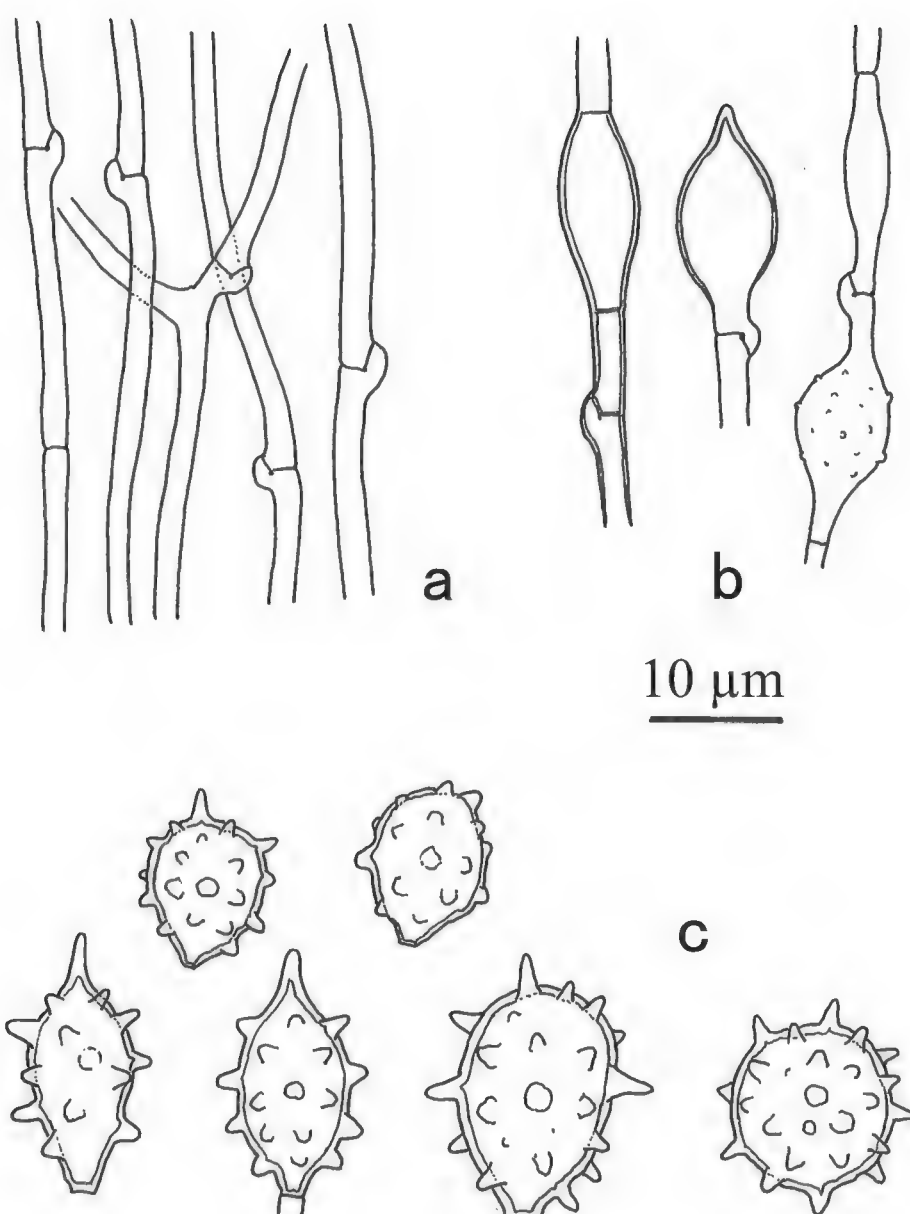


FIG. 3. Structures of *Postia stellifera* (from ex-type culture).  
 —a: Generative hyphae from advancing zone. —b: Young chlamydospores.  
 —c: Mature chlamydospores.

sordid white to grayish; pores angular, (1–)2–3 per mm, dissepiments thin, entire, with conspicuous hyphal pegs near the pore mouth. Context fleshy in fresh condition, soft and flexible in dried condition, spongy near the pileus surface, dense near the tubes; grayish brown near the pileus surface, partly light brown near the tubes, otherwise whitish to pale orange, up to 7 mm thick. Tubes whitish, fleshy in fresh condition, drying more or less brittle, decurrent on stipe, up to 5 mm deep. Stipes cylindrical, stipe surface pubescent, white in fresh condition, light brown to grayish in dried condition, up to 5 cm long and 1.5 cm wide.

Hyphal system monomitic both in context and trama. Contextual generative hyphae with clamp-connections, thin- to thick-walled with a distinct lumen,

mostly sinuous, occasionally branched, hyaline, IKI–, inflated hyphae abundant, 2–15 µm wide. Chlamydospores scattered to abundant in the context, verrucose, appendages up to 2.5 µm long, hyaline to yellow, 7.5–12.5 × 6.8–10.8 µm (excluding appendages). Tramal generative hyphae with clamp-connections, thin-walled, straight or sinuous, sparsely to conspicuously branched, hyaline, IKI–, 1.8–5 µm wide. Cystidioles present in hymenium, fusoid to mammillate, smooth, thin-walled, 14–22 × 4–5.5 µm. Basidia clavate, 4-sterigmate, with a basal clamp, 18–25 × 5–7 µm. Basidiospores oblong ellipsoid to short cylindrical, thin-walled, smooth, hyaline, IKI–, 4.5–5.5 × 1.8–2.3 µm,  $L = 5.07$  µm,  $W = 2.86$  µm,  $r = 2.21$ – $2.78$ ,  $R = 2.44$  ( $n = 23/1$ ).

**CULTURAL CHARACTERISTICS** — Growth slow, 1.2–1.4 mm/day, plates covered in 6 weeks. Advancing zone bayed, appressed, white. Mat at first white, aerial mycelium woolly to flat, becoming cream to light brown from the center. Reverse unchanged. Odor indistinctive. Hymenophore development not seen within 6 weeks. Generative hyphae from the advancing zone thin-walled, moderately branched, hyaline, 1.5–4 µm wide, with clamp-connections. Generative hyphae from aerial mycelium and submerged mycelium as in advancing zone. Chlamydospores abundant, produced intercalary or on the apex of hyphae, at first fusoid, thin-walled, smooth and hyaline, later ellipsoid to subglobose, thick-walled, distinctly verrucose to spinose, appendages up to 4 µm long, hyaline to yellow, 7–20 × 6–12 µm (excluding appendages).

**EXTRACELLULAR OXIDASE ACTIVITIES** — 1-naphthol, –; tyrosine, +.

**SPECIES CODE** — 2, 3, 7, 34, 36, 38, 46, 56 (Nobles 1965); 2, 9, 13, 15, 22, 30, 31, 39, 45, 52, (53), 85, 91 (Stalpers 1978).

**EXAMINED CULTURE** — PEN49 (ex-type strain, isolated from TFM F-20668).

**TYPE OF ROT** — unknown, but probably brown rot.

**OTHER SPECIMEN EXAMINED** — Malaysia. Perak, Taman Negara Royal Belum, alt. 259 m, on soil, 18 June 2007, leg. BK Thi (KEP FRIM4583).

## Discussion

Within the genera of polypores (Ryvarden 1991), the micro-morphological and physiological (type of rot — viz. in all probability a brown rot) features of this species would point toward *Postia* as a possible genus. In addition to the decay type, the following characteristics are common to the present fungus, *Postia* and its allies: white and fleshy to soft corky context, poroid hymenophore, monomitic hyphal system, generative hyphae with clamp-connections, and smooth basidiospores without distinct reactions in iodine reagents.

Our phylogenetic study also indicates that this fungus is related to several *Postia* species, *Amylocystis lapponica*, and *Jahnoporus hirtus*. However, the phylogenetic position of *P. stellifera* within a hypothetical *Postia* s.l. clade is still



unclear, because sequences of many *Postia*, including the type species, are still unavailable and the clade consisting of *Postia* species is weakly supported.

One of the very distinctive characteristics of *P. stellifera* is the presence of a well-developed stipe with a terrestrial habit, a feature hitherto unknown in *Postia* and other related genera. Several polypore genera accommodate only stipitate and terrestrial species such as *Albatrellus* Gray, *Coltricia* S.F. Gray, *Boletopsis* Fayod, *Cornerporus* T. Hatt., *Diacanthodes* Singer and *Polyporoletus* Snell in addition to *Jahnoporus*. However, a few genera include both lignicolous and terrestrial species, e.g., *Microporellus* Murrill with *M. clemensiae* (Murrill) Ryvarden and *M. inusitatus* (Lloyd) Corner both terrestrial versus *M. grandiporus* Corner and *M. peninsularis* (Corner) Decock, both lignicolous (Corner 1987, Decock 2001). *Phylloporia* Murrill and *Amauroderma* Murrill also accommodate both lignicolous and terrestrial species.

Another distinctive characteristic of *P. stellifera* is the presence of verrucose chlamydospores in the context, also present in culture on artificial media. Chlamydospores are present in the context of several *Postia* species such as *P. ptychogaster* (F. Ludw.) Vesterh., *P. rennyi* and *P. brunnea* Rajchenb. & P.K. Buchanan (Rajchenberg & Buchanan 1996, Ryvarden & Gilbertson 1994), and many produce subglobose to ellipsoid chlamydospores in cultures, but they are always smooth.

After intensive examination of the oxidative reactions of wood-decay fungi, Käärik (1965) listed the following 3 species that did not have laccase but had tyrosinase as in *P. stellifera*: *Lentinus lepideus* (Fr.) Fr. [= *Neolentinus lepideus* (Fr.) Redhead & Ginns], *Merulius lacrymans* (Wulfen) Schumach. [= *Serpula lacrymans* (Wulfen) J. Schröt.], and *Trechispora brinkmannii* (Bres.) D.P. Rogers & H.S. Jacks. [= *Sistotrema brinkmannii* (Bres.) J. Erikss.]. Like these three species, *P. stellifera* is in all probability also a brown rot fungus.

*Amylocystis lapponica* is characterized by amyloid cystidia and hyphae but is otherwise similar to *Postia* with monomitric hyphal system and a rot (Ryvarden & Gilbertson 1993). Nobles (1958) placed *J. hirtus* in the group positive for extracellular oxidases on the basis of the Bavendamm reaction and application of ethanol gum guaiacum. Chang (1994) also concluded that this is a white rot fungus using Bavendamm reaction. However, these methods cannot differentiate laccase and tyrosinase (Harkin et al. 1974) and are unable to evaluate the decay type.

*Jahnoporus* is the only genus to accommodate a stipitate and terrestrial species among the allied genera of *P. stellifera* and is often placed in *Albatrellaceae* (Kirk et al. 2008). Another distinctive characteristic of *Jahnoporus* is the large and spindle-shaped basidiospores that are unknown both in phylogenetically related genera and the morphologically similar genus *Albatrellus* (Gilbertson & Ryvarden 1986). We prefer not to put *P. stellifera* into *Jahnoporus* because

of the difference in basidiospore morphology and the presence of verrucose chlamydospores in the context of the former. The phylogenetic position of another species of *Jahnoporus*, *J. pekingensis* (J.D. Zhao & L.W. Xu) Y.C. Dai, is still unknown, but it also has large and more or less fusiform basidiospores that are different from those of *P. stellifera* (Dai 2003).

The present fungus may be easily mistaken for an *Albatrellus* species because of the terrestrial habit and macro-morphology, but this genus is hitherto unknown from lowland rainforest of Southeast Asia, although there are a few reports of it from the highlands of Malaysia and Papua New Guinea (Corner 1989, Quanten 1997). Most of the *Albatrellus* species are considered to be mycorrhizal and difficult to cultivate on artificial media and/or their growth is much slower (18.3–33.0 mm/8-wks on PDA, Akama et al. 2008). Most of the *Albatrellus* species have short ellipsoid to subglobose basidiospores (Gilbertson & Ryvarden 1986; Ryvarden & Gilbertson 1993) while our species has long ellipsoid basidiospores. Additionally, verrucose chlamydospores are unknown in *Albatrellus*.

Most *Albatrellus* species are included in the russuloid clade, except *A. syringae* (Parmasto) Pouzar and *A. peckianus* (Cooke) Niemelä, which are placed in ‘the residual polyporoid clade’ where other members of this clade are lignicolous and associated with a white rot (Binder et al. 2005, Bruns et al. 1998, Cui et al. 2008, Ryman et al. 2003). In addition to their phylogenetic status, the cultural characteristics suggest that *A. syringae* is possibly a white rot fungus (Niemelä 1970, Stalpers 1992), and Ryman et al. (2003) implied that it should be excluded from *Albatrellus*. *Albatrellus peckianus*, which has been reported to be attached to buried wood of *Fagus* and *Tilia* (Lowe 1942, Overholts 1953), is also possibly a saprobe. As in *A. syringae* and *A. peckianus*, *P. stellifera* is phylogenetically isolated from *Albatrellus* sensu stricto, despite their macro-morphological similarity.

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## A new record of *Gliocephalotrichum simplex* from India

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**Abstract** — During a survey of interesting and rare fungi infecting economically important plants in the forests of the Western Ghats in India, an uncommon fungal species was isolated from fruits of *Terminalia chebula*. The fungus has distinctive morphological features such as a whorl of sterile arms subtending penicillate branches bearing yellowish masses of elongated to ellipsoidal conidia. Based on morphological characters and a comparison of sequences of the internal transcribed spacer region of rDNA (ITS1-5.8S-ITS2), the fungus was determined to be *Gliocephalotrichum simplex*, a species not previously known from India.

**Key words** — anamorph, fungal diversity, *Hypocreales*, ITS sequence

### Introduction

India is a tropical country that harbors considerable fungal biodiversity (Bilgrami et al. 1991, Jamaluddin et al. 2004). As part of our ongoing effort to discover and preserve fungi, we are making regular surveys and isolating rare and unusual fungi. During 2008–09 partially degraded fruits of *Terminalia chebula* were collected from the forest floor in Western Ghats in Maharashtra state, India. From these fruits a fungus was isolated and identified as *Gliocephalotrichum simplex*. This fungus has never been recorded from India. The present communication describes this fungus from India on artificial media isolated from fruits of *T. chebula*.

### Materials and methods

#### Isolation, pure culture and microscopic examination

Samples of *Terminalia* fruits were collected in separate paper bags and transported to the laboratory. The fruit samples were surface-sterilized by dipping in 70% ethanol for 10 min, then rinsed in distilled water and incubated in a moist chamber at  $25 \pm 2^\circ\text{C}$ . A yellowish to grayish fungal growth appeared on the fruit surface after 4 days. Direct

streak and serial dilution plate methods were used to isolate the fungus as a pure culture. Potato dextrose agar (potatoes, peeled, sliced 200 g, dextrose 20.0 g, agar 20 g, water 1L) and V8 (HIMEDIA) were used as the isolation medium. The isolation plates were sealed with parafilm (M250-HIMEDIA) and incubated at ambient lab temperature (25°C).

A Nikon trinocular stereozoom microscope (Model SMZ- 1500 with Digi CAM) was used for direct observation of the fungal growth pattern on the fruit surface. For microscopic details and photomicrographs, an Olympus CX-41 microscope was used. Specimens were mounted in lactophenol-cotton blue and distilled water for microscopic studies. Measurements of fungal structures were made with an ocular micrometer.

The specimen is deposited in Ajrekar Mycological Herbarium (AMH, according to Holmgren et al. 1990) and a pure culture is deposited in the National Fungal Culture Collection of India (NFCCI-WDCM 932), MACS' Agharkar Research Institute, Pune, India.

### DNA isolation

The fungal strain was maintained on PDA slants. DNA was extracted from cultures grown on PDA plates for two weeks at 28°C by first homogenizing the mycelium in FastPrep®24 tissue homogenizer (MP Biomedicals GmbH, Germany) and then using the CTAB method of Graeser et al. (1999).

### PCR amplification

For ITS-PCR the universal primers ITS4 (5' TCC TCC GCT TAT TGA TAT GC3') and ITS5 (5' GGA AGT AAA AGT CGTAAC AAG G 3') amplifying a DNA fragment of about 700 bp of the rDNA gene were used (White et al. 1990). The PCR mixture contained reaction buffer (10 mM TrisHCl pH 8.0, 50 mM KCl, 1.5 mM MgCl<sub>2</sub>), 200 µM of each deoxynucleoside triphosphates (Genei, Bangalore, India), 50 pM each of primers, 1U of Taq polymerase (Genei, Bangalore, India), and 25 ng of template DNA. Samples were overlaid with sterile mineral oil and amplified through 30 cycles in a thermocycler (Eppendorf MastercyclerAG, Hamberg, Germany) as follows: initial denaturation for 5 min at 95°C, denaturation for 1 min at 95°C, annealing for 1 min at 56°C, and extension for 1 min at 72°C. This was followed by a final extension step for 10 min at 72°C. The resulting PCR product was checked on 1.2% agarose gel (Sigma).

### Sequencing

PCR products were cleaned with Axygen PCR cleanup kit (Axygen Scientific Inc, CA, USA) and sequenced using primers ITS4 and ITS5 (White et al. 1990) on an automated DNA Sequencer ABI 3130 (Applied Biosystems, USA).

### Sequence Alignment & Phylogenetic tree

rDNA sequences (ITS1-5.8S-ITS2) of the Indian isolate of *G. simplex* were manually aligned with those of known *G. simplex* sequences and the other six species of *Gliocephalotrichum* in the NCBI database (Table 1) using text editor option of the software MEGA for similarity. The manually edited sequence of NFCCI1496 was deposited in the EMBL nucleotide sequence database (FN550111) and was also subjected to a BLAST search. The neighbor-joining tree was derived from analyses of ITS1-5.8S- ITS2 sequences using Mega4.0 software.

### Taxonomic description

*Gliocephalotrichum simplex* (J.A. Mey.) B.J. Wiley & E.G. Simmons,

Mycologia 63(3): 578, 1971.

FIGS 1–8

HABITAT: On rotting fruit of *Terminalia chebula* Retz. (*Combretaceae*).

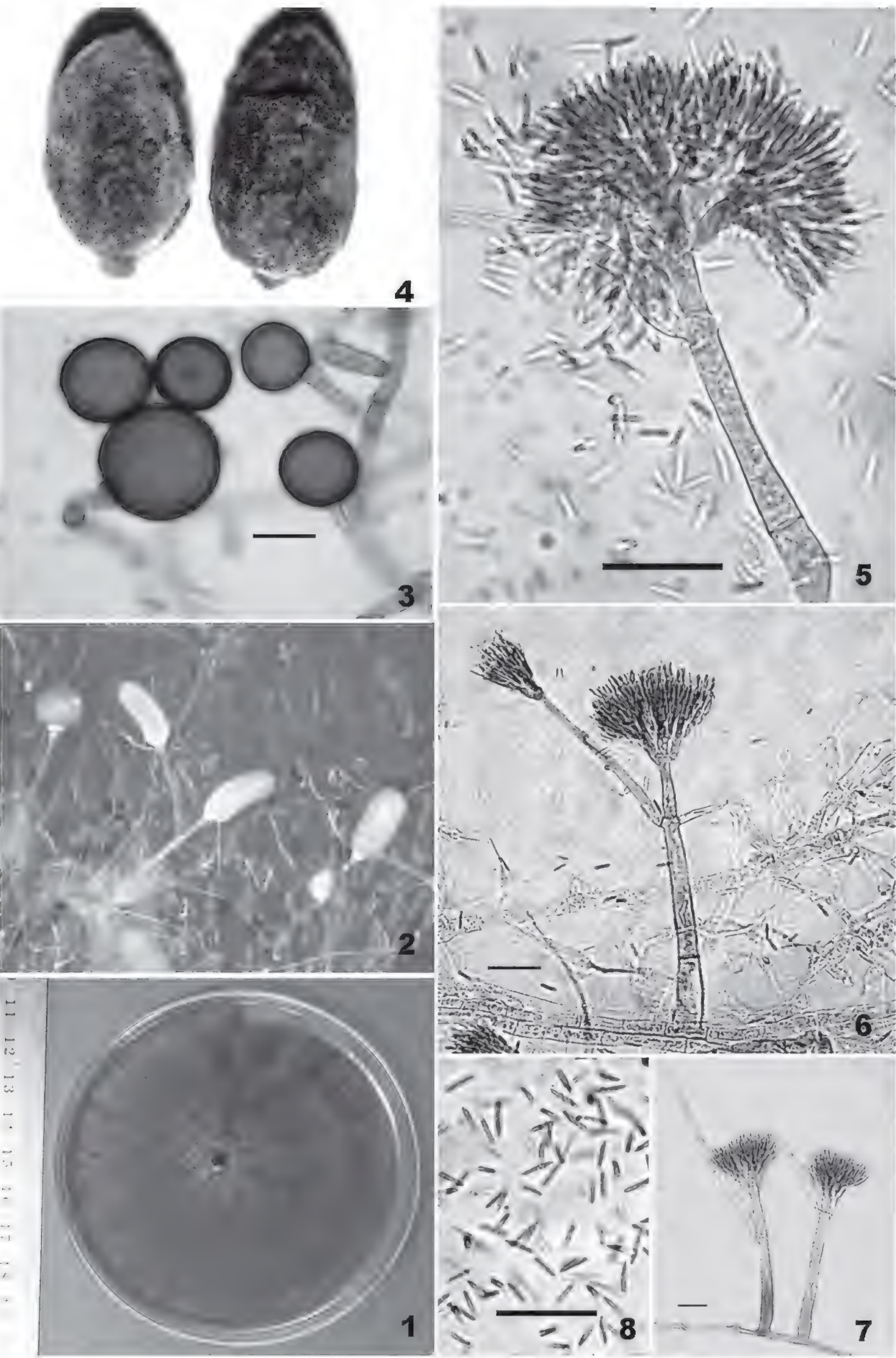
TELEOMORPH: Unknown.

ANAMORPH: Optimum temperature for growth 25–28°C. Colony radius after 3 d on PDA (80 mm), CMA (75 mm) and V8 (70 mm). Colonies on PDA off-white in centre, floccose cottony, buff, golden brown, sporulating, margin irregular. Reverse buff. Appearance in nature: Substrate brown to blackish, covered with grayish-white colonies that later turn yellowish and spread over entire outer surface. Hyphae branched, septate, hyaline, smooth, 7.5–10.5 µm wide. Chlamydospores one-celled, terminal to intercalary or lateral, subglobose to mostly globose, thick-walled, golden brown with short stalk, 20–35 × 20–32 µm diam. Sterile hairs 1–2, originating from branching point of conidiophores or beneath septum subtending penicillus, hairs hyaline 3–8 septate, 125–412 µm long, base broad, tip narrow. Conidiophores erect, simple to branched, arising directly from submerged mycelium, hyaline to subhyaline, 80–162.5 × 7.5–10 µm, broad at base gradually narrower towards apex, 2–6-septate, at apex bearing a compact penicillus, with slimy head. Penicillus of successive branches, primary branches 7–10 × 4–6 µm, secondary branches 6–8 × 4–5 µm, tertiary branches 5–7 × 4–5 µm, quaternary branches 5–6 × 2–4 µm. Conidia cylindrical to ellipsoidal, smooth, hyaline, 7.5–9(–10) × 1–1.5 µm.

SPECIMEN EXAMINED: India, Mahabaleshwar (17°55'15"N 73°39'21"E), Maharashtra, on degraded fruits of *Terminalia chebula* (*Combretaceae*), Oct. 2008, L. S. Yadav, AMH 9279, Culture No. NFCCI1496

NOTES—The genus *Gliocephalotrichum* J.J. Ellis & Hesselt., typified by *G. bulbilium* J.J. Ellis & Hesselt., is mainly characterized by the origin of the sterile arms and the conidia along with the morphology and dimension of chlamydospores (Ellis & Hesseltine 1962, Decock et al. 2006). This genus has been expanded to include six additional species: *G. bacillisporum* Decock & Huret, *G. cylindrosporum* B.J. Wiley & E.G. Simmons, *G. longibrachium* Decock & Charue, *G. microchlamydosporum* (J.A. Mey.) B.J. Wiley & E.G. Simmons, *G. ohiense* L.H. Huang & J.A. Schmitt, and *G. simplex* (Ellis & Hesseltine 1962, Wiley & Simmons 1971, Huang & Schmitt 1973, Decock et al. 2006).

Sequencing of rDNA (ITS1, ITS2 and 5.8S) shows that our isolate is *Gliocephalotrichum simplex*, a species not previously recorded from India. The present strain NFCCI 1496 is part of the clade formed by other strains of *G. simplex* (Fig. 9), however, it differs slightly from its closest strain MUCL46551 from Singapore by three nucleotide positions, i.e. two transition of C→T at 202 and 316 bases along with an insertion of an A at position 8 (Fig. 10).





Initially, morphological differences viz. number and size of sterile arms and branched conidiophores produced on different media showed slight variation in morphological features from *G. simplex* (Wiley & Simmons 1971), but rDNA sequence comparisons showed that our isolate is indeed *G. simplex*. The setae of our isolate originate directly below the penicillus unlike the descriptions of this species. *Gliocephalotrichum simplex* is distinguished by the presence of 1–3 sterile hairs originating from 10–15 µm below the penicillus and cylindrical conidia measuring 7.5–9(–10) × 1–1.5 µm (n = 100 spores) accommodate this isolate in *G. simplex* (Wiley & Simmons 1971).

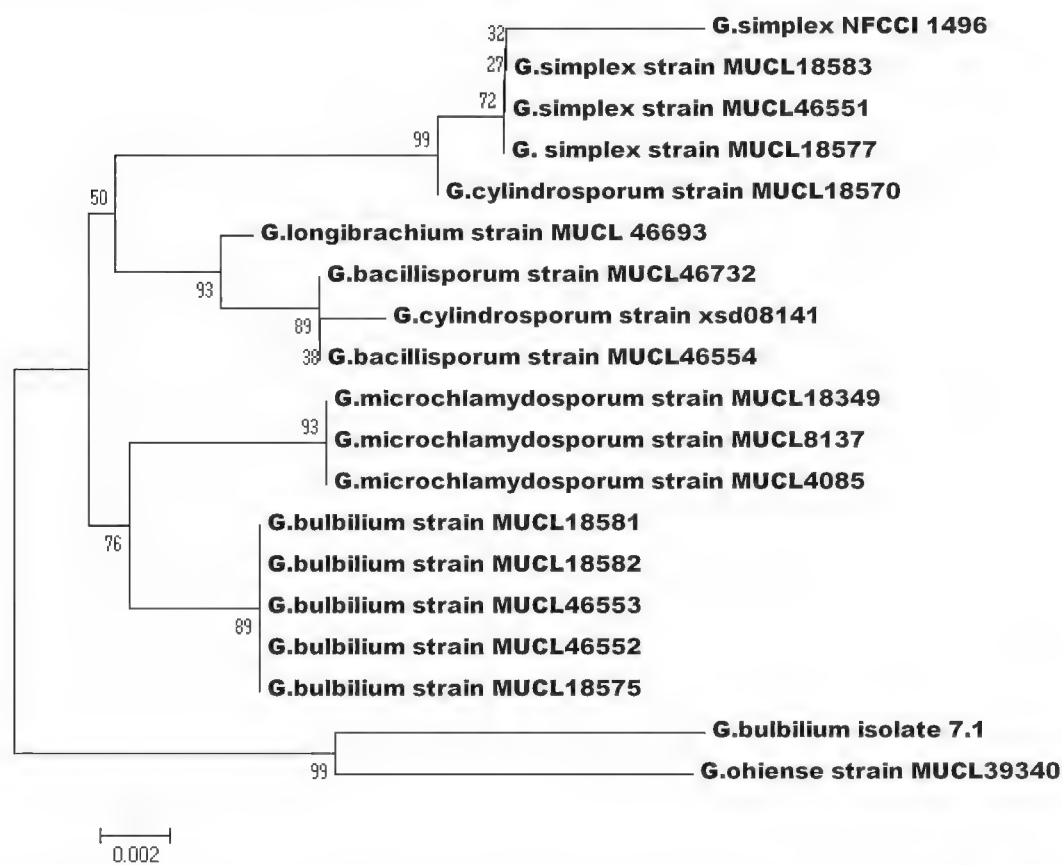


FIG. 9. Neighbour joining tree based on ITS1-5.8S-ITS2 sequences showing the relationships among 21 *Gliocephalotrichum* strains representing 7 species.

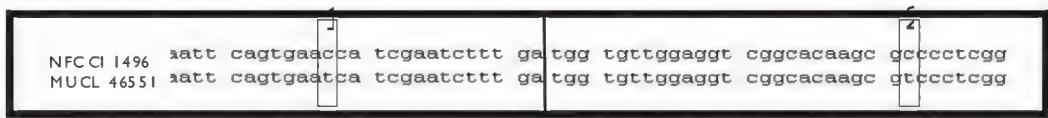


FIG. 10. Inversion (C to T) noted at two locations in *Gliocephalotrichum simplex* MUCL46551 from Singapore in comparison to NFCCI-1496 from India: position 1-base 202 (part of 5.8S gene); position 2 - base 316 (part of ITS2 region of rDNA).

FIGS. 1–8 (left). *Gliocephalotrichum simplex*. 1. Colony on PDA after 5 days. 2. Stereoscopic view of yellowish conidial heads. 3. Thick walled, golden brown, globose chlamydospores. 4. Infected fruits of *Terminalia chebula* 5. Mature conidiophores with penicilli. 6. Conidiophores with fertile stipe extensions. 7. Conidiophores with sterile arms (setae). 8. Conidia. Scale bars = 20 µm.



TABLE 1. Comparison of the rDNA sequences (ITS1-5.8S-ITS2) among isolates of *Gliocephalotrichum*.

SPECIES	STRAIN ACCESSION §	SIMILARITY *	GENEBANK/ EMBL ACC.
<i>G. simplex</i>	NFCCI 1496	-	FN550111
<i>G. simplex</i>	MUCL 46551	99%	DQ366704
<i>G. cylindrosporum</i>	MUCL 18570	98%	DQ366705
<i>G. bacillisporum</i>	MUCL 46554	97%	DQ374408
<i>G. longibrachium</i>	MUCL 46695	97%	DQ278422
<i>G. bulbilium</i>	MUCL 18582	96%	DQ381952
<i>G. microchlamydosporum</i>	MUCL 18349	96%	DQ366701

§ NFCCI: National Fungal Culture Collection of India, Pune, India;  
MUCL: Mycotheque de l’Universite Catholique de Louvian, Louvain-la-Neuve, Belgium.  
\* with NFCCI 1496

SOURCE: NCBI (<http://www.ncbi.nlm.nih.gov/>)

There is no previous record of *G. simplex* from India (Bilgrami et al. 1991, Jamaluddin et al. 2004). Earlier records of *G. simplex* from various parts of the world are mainly from soil and debris (Watanabe & Nakamura 2005), although it has been reported on fruit of rambutan (Nishijima et al. 2002). The isolate from India is reported for the first time from fruits of *Terminalia chebula*, a plant that has been used as a traditional medicine. Therefore, the present fungus is documented here as new record from India.

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# MYCOTAXON

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## Two new records of *Mucorales* from the Brazilian semi-arid region

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**Abstract** — *Apophysomyces elegans* and *Mycotypha microspora* are recorded for the first time in Brazil based on isolates from semiarid soil in the Northeast part of the country.

**Key words** — *Zygomycetes*, *Mucoromycotina*, taxonomy

### Introduction

*Apophysomyces* and *Mycotypha* belong to the subphylum *Mucoromycotina* (Hibbett et al. 2007), family *Mucoraceae*, order *Mucorales* (Benny 2005). *Apophysomyces* was first described by Misra et al. (1979), and the description of *A. elegans* (the monotype) was based on two specimens isolated from soil. This species typically produces a pyriform, apophysate multispored sporangia developed on a sporangiophore with a funnel-shaped to bell-shaped apophysis. *Apophysomyces elegans* has also been reported as an agent of zygomycosis in immunocompromised patients (Kimura et al. 1999; Liang et al. 2006; Chakrabarti et al. 2008; Reddy et al. 2008).

*Mycotypha* was introduced by Fenner (1932), who described a single species, *M. microspora*. Six species have been included in the genus, but Benny et al. (1985) accepted only three as true *Mycotypha* species. *Mycotypha microspora* was isolated as a contaminant on a plate culture of a pathogen of bitter orange (*Citrus aurantium*) and was first classified in *Mucoraceae*. Since then, *Mycotypha* has been placed in the *Choanephoraceae* (Bessey 1950) and the *Cunninghamellaceae* (Hesseltine 1952). Novak & Backus (1963) described *M. africana*, which produces zygospores with a typically mucoraceous form. Young (1969), based on the electron and phase-contrast microscopy of spores, reported that *Mycotypha* should be included in the *Thamnidaceae*. Later,

Benny et al. (1985) proposed the family *Mycotyphaceae*, including a new species *M. indica*. *Mycotypha microspora* is characterized by sporophores terminating in a mostly cylindrical fertile vesicle bearing dimorphic sporangiola subtended by conical denticles. Yeast-like budding cells and thin-walled chlamydospores are also characteristics of this species.

The purpose of this manuscript is to report the first occurrence of *Mycotypha microspora* and *Apophysomyces elegans* in Brazil. For *M. microspora* this also represents the first record for South America.

### Materials and methods

Soil samples were collected at Belém de São Francisco (8°33'59"S, 38°49'59"W) and Triunfo (7°52'28"S, 38°06'03"W), located in the semi-arid region of the State of Pernambuco, Northeastern Brazil. Belém de São Francisco is characterized by xerophilous vegetation with patches of deciduous forest. The typical biome is named caatinga and the climate is tropical semi-arid. Triunfo comprises semi deciduous forest and, according to Koeppen's classification, the climate is hot and humid tropical. Both areas are included in the Brazilian semi-arid region, which covers more than 969,589 km<sup>2</sup> (Ministério da Integração Nacional 2005).

The samples of soil were collected with a sterilized spatula, placed in plastic bags and taken to the laboratory. Soil particles (5 mg) were placed on sets of Petri dishes containing MEYE (Benny 2008) plus chloramphenicol (100mg/L). The plates were left on a bench at room temperature (28 ± 2°C) under light and dark periods for 72 hours. Fragments of mycelium were removed directly from the samples at the stereomicroscope and transferred to Petri dishes with M agar (O'Donnell 1979). Identification and descriptions were based on macroscopical (color, aspect and diameter of the colonies) and microscopical (microstructures) characters according to Benny & Benjamin (1976) and Misra et al. (1979).

### Taxonomy

*Apophysomyces elegans* P.C. Misra, K.J. Srivast. & Lata, Mycotaxon 8(2): 377 (1979)

FIG. 1 A–D

SPECIMEN EXAMINED: Brazil, Pernambuco, Triunfo, soil, Jan. 2010, A.L.C.M.A. Santiago (URM-Culture collection 6169).

Colonies remaining white on M agar, reverse pale yellow, 9 cm diam in 72 hour at 28°C. SPORANGIOPHORES growing slowly, after 7 days, often single, developing at right angles from aerial stolon-like hyphae which generally becomes delimited by two septa near the place of origin of the sporangiophore; erect, unbranched, thick-walled, smooth, light brown, becoming darker near the base and darker and thicker below the apophyses, up to 550 µm long and 5 µm wide near the base. SPORANGIA hyaline at first, becoming light yellowish-brown, terminally, pyriform, distinctly apophysate, 20–50 µm diam. APOPHYSES funnel-shaped to bell-shaped, 12–47 µm high and 17–27.5 µm diam at the



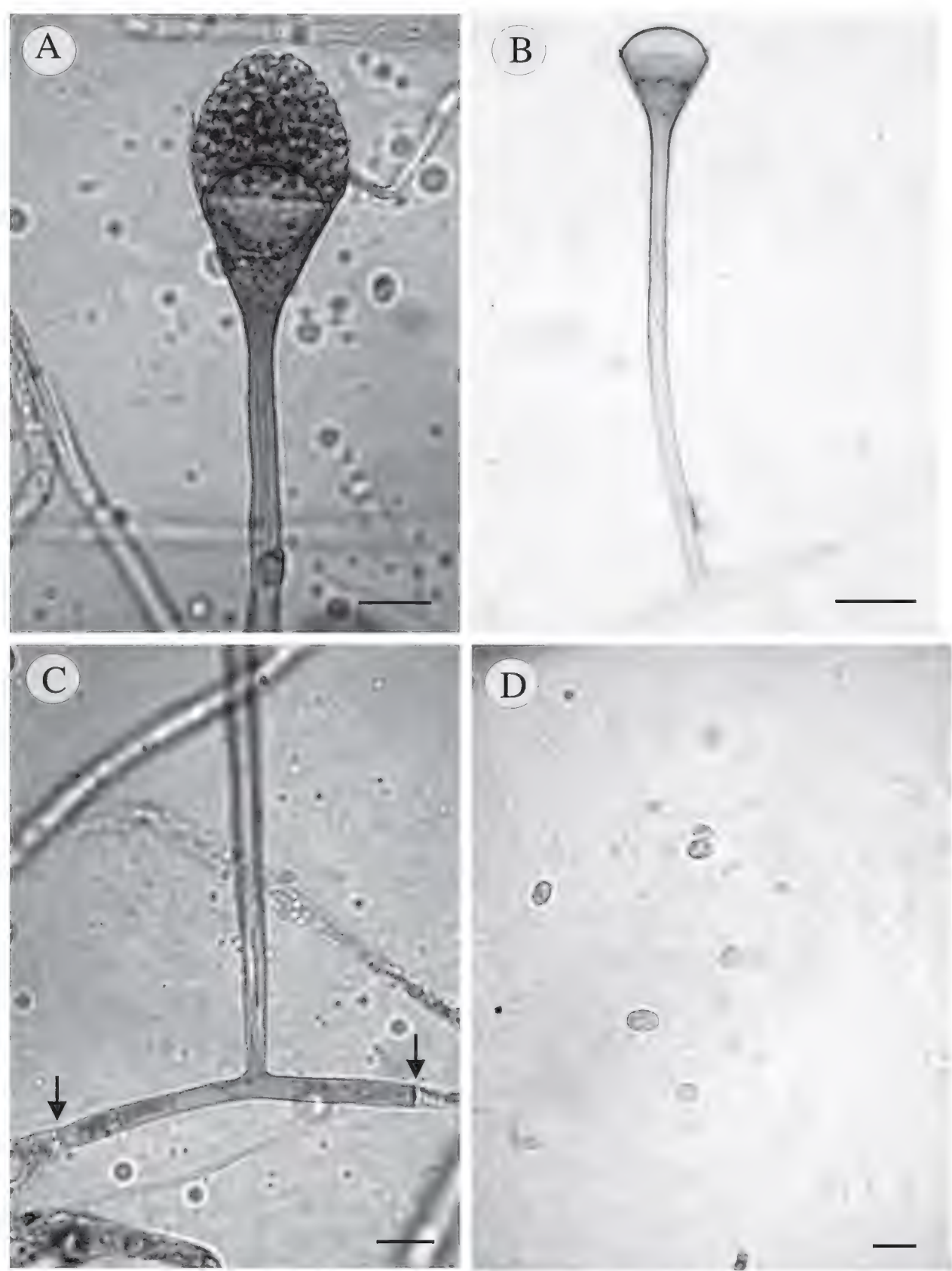


FIG. 1 *Apophysomyces elegans*. A) Sporangiophore with sporangium; B) Sporangiophore with funnel-shaped apophysis and columella; C) Stolon-like hypha delimited by two septa near the place of origin of the sporangiophore; D) Sporangiospores. Scale bars: A, C, D = 10µm, B = 20 µm.

widest part; smooth-walled, light brownish. COLUMELLAE hemispherical, thin-walled, subhyaline, 20–30 µm diam, collar distinct. SPORANGIOSPORES oblong,

sometimes subglobose, subhyaline, minutely roughened,  $4.5\text{--}8.5(-12.5) \times 4\text{--}5.5(-6.5) \mu\text{m}$ . RHIZOIDS unbranched, subhyaline. ZYGOSPORES not observed.

HABITAT: Soil

GEOGRAPHIC DISTRIBUTION: Australia (Cooter et al. 1990), Caribbean (Meis et al. 1994), Colombia (Ruiz et al. 2004), India (Mirza et al. 1979; Lakshmi et al. 1993; Shakrabarti et al. 2003) and USA (Blair et al. 2002; Liang et al. 2006; Ferguson et al. 2007).

REMARKS: The characteristics of the *Apophysomyces elegans* strains reported here show a close similarity with the original description of Misra et al. (1979). However, differences in colony color and sporangiospore walls were observed. The colonies were persistently white, as also described by Lakshmi et al. (1993), but Misra et al. (1979) and Cooter et al. (1990) reported colonies as white at first, becoming brownish-gray, and then creamy white to buff with age. Recently, Reedy et al. (2008) described the colonies as initially white, turning brownish-gray or yellow. The fact that different authors have used dissimilar culture media for descriptions may explain this variation of color. Curiously, the *A. elegans* sporangiospores described here are minutely roughened, differing from the smooth ones reported by Misra et al. (1979). However, we did not consider these differences enough to characterize a new taxon. *Apophysomyces elegans* has some microscopic features similar to those of species of *Absidia*, like sporangiophores arising from stolons and pyriform, apophysate sporangia. Nevertheless, *Apophysomyces* differs from *Absidia* in bearing a more pronounced, funnel-shaped to bell-shaped, apophysis. In addition, the sporangiophore wall below the apophyses is dark and thick in *Apophysomyces* (Mirza 1979; Lakshmi 1993).

*Mycotypha microspora* Fenner, Mycologia 24: 196 (1932)

FIG. 2 A–D

SPECIMEN EXAMINED: Brazil, Pernambuco, Belém de São Francisco, soil, Jan. 2010, A. L. C. M. A. Santiago (URM-Culture collection 6170).

Colony with limited growth after 15 days at 28°C in M agar; more or less zonate, later deep gray or brown with age. SPOROPHORES simple at first, some secondarily branched, hyaline at first, becoming grayish brown in age, irregularly multiseptate, particularly below the VESICLE, 3 mm high,  $3\text{--}18.5 \mu\text{m}$  diam. FERTILE VESICLES terminal, mostly cylindrical, rounded at the apex, appearing minutely roughened, bearing sporangiola over entire surface, except at extreme tip,  $20\text{--}580 \times 10\text{--}40 \mu\text{m}$ . SPORANGIOLA dimorphic, forming two different layers over surface of vesicle; at outer layer, ovoid to obovoid,  $4\text{--}6 \times 3\text{--}5 \mu\text{m}$ , pale bluish-gray, smooth, globose to subglobose borne on conical pedicels; at inner layer,  $3\text{--}5.5 \mu\text{m}$  in diam, pale bluish-gray, smooth, born on conical pedicels. After dehiscence, the sporangioles bear remnant of pedicel. ZYGOSPORES not observed.

HABITAT: Soil

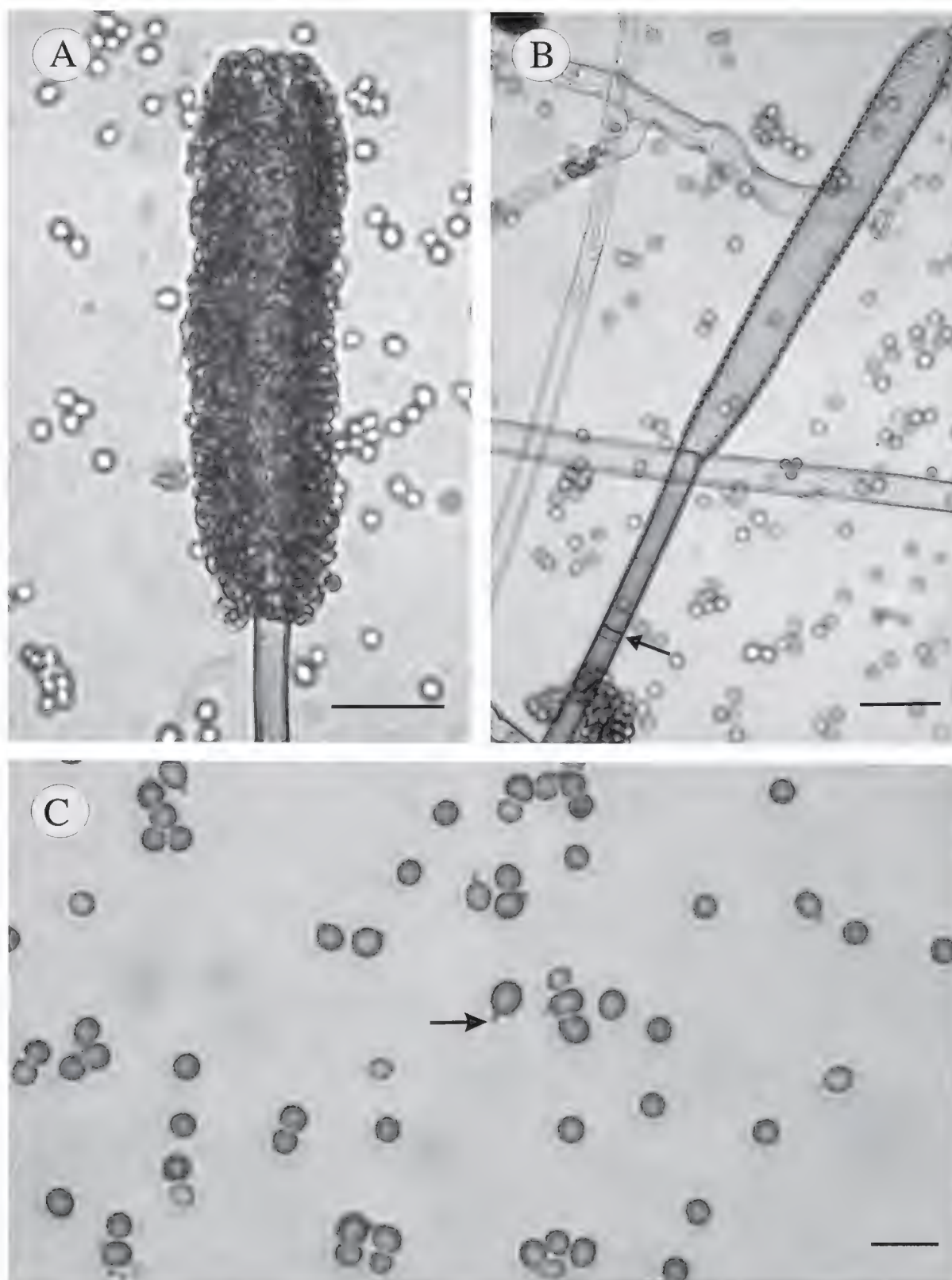


FIG. 2 *Mycotypha microspora*. A) Sporangiophore with terminal fertile vesicle and sporangia; B) Terminal fertile vesicle after dehiscence of the sporangia; septa produced near the vesicle. C) Globose and ovoid to obovoid sporangia with remnant of pedicels.  
Scale bars: A, B = 30  $\mu$ m; C = 10  $\mu$ m.

GEOGRAPHIC DISTRIBUTION: Belgium (IHEM), Finland (IMI), France (Lacroix et al. 2007; IHEM), Germany (IMI), Great Britain (IMI), India (Ray & Mukerji 1961), Japan (NBRC), Libya (IMI), Netherlands (CBS), Nigeria (IMI), Poland (IMI), USSR (CBS), Thailand (CBS), Turkey (MUCL), USA (Benny & Benjamin 1976).

REMARKS: The strain characteristics of *M. microspora* reported here are very close to the original description by Benny & Benjamin (1976). The known species of *Mycotypha* are morphologically similar, but *M. microspora* differs from *M. africana* in producing ovoid to obovoid external sporangiola, while in the latter the external sporangiola are cylindrical. In *M. microspora* the septa in the sporophore are usually produced near the apex but may also be formed near the base, while in *M. indica* the septa are only produced near the base (Benny et al. 1985).

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## MYCOTAXON

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***Sphaerodes* mycoparasites  
and new *Fusarium* hosts for *S. mycoparasitica***

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**Abstract** — A comprehensive key, based on asexual stages, contact mycoparasitic structures, parasite/host relations, and host ranges, is proposed to distinguish those species of *Sphaerodes* that are biotrophic mycoparasites of *Fusarium*: *S. mycoparasitica*, *S. quadrangularis*, and *S. retispora*. This is also the first report of *S. mycoparasitica* as a biotrophic mycoparasite on *Fusarium culmorum* and *F. equiseti* in addition to its other reported hosts (*F. avenaceum*, *F. graminearum*, and *F. oxysporum*). In slide culture assays, *S. mycoparasitica* acted as a contact mycoparasite of *F. culmorum*, and *F. equiseti* producing hook-like attachment structures. Fluorescent and confocal laser scanning microscopy showed that *S. mycoparasitica* is an intracellular mycoparasite of *F. equiseti* but not of *F. culmorum*. All three mycoparasitic *Sphaerodes* species were observed to produce asexual (anamorphic) stages when challenged with *Fusarium*. Furthermore, a phylogenetic tree, based on (large subunit) LSU rDNA sequences, depicted closer relatedness to one another of these *Fusarium*-specific *Sphaerodes* taxa than to the non-mycoparasitic *S. compressa*, *S. fimicola*, and *S. singaporensis*.

**Key words** — ascomycete, coevolution

**Introduction**

Mycoparasitism refers to the parasitic interactions between one fungus (parasite) and another fungus (host). These relationships can be categorized as either necrotrophic or biotrophic (Boosalis 1964; Butler 1957). Differences between necrotrophic and biotrophic mycoparasites were reviewed and outlined by Jeffries & Young (1994). This paper emphasizes biotrophic *Sphaerodes* Clem. (*Ascomycota*) mycoparasites and their association with fungi, in particular *Fusarium* Link. Biotrophic mycoparasitic ascomycete and basidiomycete fungi are characterized by intimate contact with host cells (Bauer & Oberwinkler 2004; Gams et al. 2004), with or without penetration. This intimate contact involves generation of short haustoria and appressoria or absorptive mycoparasitic cells.

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\* Corresponding author

The cytoplasm of the host hyphae remains healthy in at least some phase(s) of mycoparasitic interactions (Jeffries 1995).

Among pyrenomycetous orders, *Melanosporales* contains the largest number of biotrophic mycoparasites (Davey et al. 2008; Zhang et al. 2002), mainly within *Melanospora* Corda, *Persiciospora* P.F. Cannon & D. Hawksw., *Sphaerodes*, and *Syspastospora* P.F. Cannon & D. Hawksw. (Cannon & Hawksworth 1982; Harveson & Kimbrough 2001; Posada et al. 2004). *Sphaerodes* is a relatively small genus with unique morphological features to some extent similar to *Melanospora* and *Microthecium* Corda (García et al. 2004). Interestingly, most of the known *Sphaerodes* mycoparasitic taxa associate with *Fusarium* species — causal agents of diseases in plants and toxicosis in humans and animals (Goh & Vujanovic 2010; Harveson & Kimbrough 2001; Vujanovic & Goh 2009). To distinguish *Sphaerodes* from other genera in *Melanosporales*, ascospore characters such as wall ornamentation and shape are utilized (Zhang et al. 2002).

Identification of *Sphaerodes* species is mostly based on morphological attributes of their ascomata, structural details of ascomatal wall and neck tissues, as well as distinctive ascospore shape and ornamentation. To date, their anamorphs and their mode of mycoparasitism of *Fusarium* are poorly known.

Among all the described *Sphaerodes* species, five have been reported associated with fungal hosts (Farr & Rossman 2009). *Sphaerodes mycoparasitica* Vujan., *S. quadrangularis* Dania García, Stchigel & Guarro and *S. retispora* (Udagawa & Cain) P.F. Cannon & D. Hawksw. var. *retispora* were reported to be biotrophic mycoparasites of *Fusarium* species (Vujanovic & Goh 2009; Goh & Vujanovic 2010; Harveson & Kimbrough 2001), whereas *S. episphaeria* (W. Phillips & Plowr.) Clem. was associated with *Hypomyces* sp. (Cannon et al. 1985). *Sphaerodes retispora* var. *retispora* was the first *Sphaerodes* species reported to be a biotrophic mycoparasite of *Fusarium oxysporum* (Harveson & Kimbrough 2001). Recently, *S. mycoparasitica* and *S. quadrangularis* were also observed to establish biotrophic mycoparasitic relationships with a few *Fusarium* taxa, including red-pigmented species such as *F. avenaceum* and *F. graminearum* (Goh & Vujanovic 2010; Vujanovic & Goh 2009). However, there is no single report comparing these three *Sphaerodes* biotrophic mycoparasites, specific to *Fusarium*, in terms of differences in mycoparasitic contact structures, host ranges, and anamorphic reproductive structures.

Therefore, the purpose of this paper is to document two new *Fusarium* hosts for *S. mycoparasitica*, as well as to discuss and describe differences in these three biotrophic mycoparasites based on parasitic contact structures, phialidic stages and host ranges. Furthermore, a phylogenetic analysis based on LSU (large subunit) rDNA is incorporated into this study to determine the role of host specialization in the evolution of mycoparasitic *Sphaerodes*.

## Materials and methods

### Fungal isolates and growth

*Sphaerodes mycoparasitica* was first isolated and described by Vujanovic & Goh (2009) as an obligate biotrophic mycoparasite of various *Fusarium* taxa from Canadian agricultural fields. *Sphaerodes quadrangularis* (CBS112764 strain) was first reported as a facultative biotrophic mycoparasite of *Fusarium avenaceum*. *Sphaerodes retispora* var. *retispora* (CBS 994.72), isolated from Japanese soil, was also obtained from Centraalbureau voor Schimmelcultures (CBS, Fungal Biodiversity Centre) Baarn, The Netherlands. Biotrophic mycoparasite *Sphaerodes mycoparasitica* SMCD2220 and pathogenic *Fusarium* strains (*F. arthrosporioides* SMCD2247, *F. culmorum* SMCD2248, *F. equiseti* SMCD2134, *F. flocciferum* SMCD2135, *F. poae* SMCD2136, and *F. torulosum* SMCD2139) were obtained from the Saskatchewan Microbial Collection and Database (SMCD), Saskatchewan, Canada. All fungal isolates were grown and maintained on potato dextrose agar (PDA) (Difco, BD, Sparks, Maryland) prior to the study.

### Fungal-fungal interactions

For examination of the interaction between isolates of *Sphaerodes* and *Fusarium* species, both biotrophic mycoparasite and *Fusarium* isolates were inoculated and assessed using slide culture assays proposed in Cole & Kendrick (1968) and Jacobs et al. (2005), with slight modifications as in Goh & Vujanovic (2010). Slides were maintained in a sterile humidity chamber as outlined in Kavková & Čurn (2005) and daily observations on the hyphal interactions at the meeting place (contact zone) were performed under a Carl Zeiss Axioskop2 equipped with Carl Zeiss AxioCam ICc1 camera with 20×, 40× and 100× objectives. Formation of biotrophic mycoparasitic contact structures attaching *Sphaerodes* species to *Fusarium* hyphae were examined, recorded, and compared to drawings from the literature (Jordan & Barnett 1978; Rakvidhyasastra & Butler 1973; Whaley & Barnett 1963). Diameters of both parasitized and non-parasitized *Fusarium* hyphal cells were measured under light microscopy with a 100× objective lens. Each treatment used six replications consisted of *Sphaerodes* or *Fusarium* alone, and *Sphaerodes*–*Fusarium* co-inoculated. The experiment was repeated twice. In the slide-culture assay, *Fusarium* mycelia infected with *Sphaerodes* haustoria were stained with lactofuchsin (Carmichael 1955). Stained hyphae of both *Fusarium* and *Sphaerodes* in slide-culture were then examined with a Carl Zeiss Axioskop2 fluorescent microscope attached to Carl Zeiss AxioCam ICc1 with 40× and 100× objectives. Slide-culture assays were also subjected to Zeiss META 510 confocal laser scanning microscopy (CLSM) analysis to observe intracellular mycoparasitism under a C-Apochromat 63× N.A.1.2 phase-contrast water immersion objective through Z-stacking mode to scan through the *Fusarium* hyphae with intracellular infection (CLSM with 514nm excitation – argon and LP585 emission filters) (Abdellatif et al. 2009).

### Fungal morphology and taxonomy

The anamorphic stages of three mycoparasitic *Sphaerodes* species (*S. mycoparasitica*, *S. quadrangularis*, and *S. retispora* var. *retispora*) were compared in the presence of *Fusarium* hosts. Diameters of base and neck of monophialides were measured and base-

neck ratios were calculated. Genomic DNA of *S. retispora* var. *retispora* CBS 994.72 was extracted, amplified, and sequenced as outlined in Vujanovic & Goh (2009) by targeting LSU rDNA fragments with LS1/LR5 primers (Hausner et al. 1993; Rehner & Samuels 1995; Zhang & Blackwell 2002). The LSU sequence from this study and sequences retrieved from GenBank were aligned using Clustal X software (version 1.82) (Thompson et al. 1997), and edited in BioEdit (Hall 1999). Distance trees were generated with Phylogenetic Analysis Using Parsimony (PAUP) 4.0b10 software (Swofford 2000) using neighbor-joining approach, and validated using bootstrap analyses with 1000 repetitions. A fungal distance tree was prepared with sequences showing bootstrap values higher than 50%. The LSU sequence from *Sphaerodes retispora* var. *retispora* was submitted to GenBank as GU205261.

### Statistical analysis

The difference in diameters of parasitized and non-parasitized hyphal cells was analyzed with a T-test (SPSS 1990).

## Results

### Fungal-fungal interaction

Hyphae-hyphae interactions and contact structures in the contact zone were examined for seven days. On day three, *Sphaerodes mycoparasitica* was found to produce hook-shaped contact structures on *Fusarium equiseti* and *F. culmorum* (FIG. 1). On day five, more hook-shaped contact structures and intracellular penetration of *F. equiseti* were observed (FIG. 2A, 3A–D). The combination of lactofuchsin dye and fluorescent or confocal laser scanning microscopy revealed that the parasitized or penetrated *Fusarium* cells became empty (loss of cytoplasm = no fluorescence) or fluoresced with low intensity (very pale) (FIG. 3A–D) as compared to healthy *Fusarium* cells. During the seven days of observation, no *S. mycoparasitica* hyphae were observed within *F. culmorum* cells. *Sphaerodes mycoparasitica* produced hook-shaped contact structures (FIG. 1A, a) more frequently than clamp-like contact structures (FIG. 1B, b) on both *F. equiseti* and *F. culmorum*. Diameters of *F. equiseti*, but not *F. culmorum*, hyphae parasitized by *S. mycoparasitica* were observed to be significantly reduced compared to non-parasitized *Fusarium* hyphae (with T-test,  $P = 0.001$  and  $P > 0.05$ , respectively) (FIG. 4).

None of the *Fusarium* taxa tested appeared to be suitable hosts for mycoparasitic *S. quadrangularis* and *S. retispora*, even after 10 days of co-inoculation on slide cultures. No contact biotrophic parasitic structures or intracellular parasitism by *S. quadrangularis* and *S. retispora* on the tested *Fusarium* strains were observed at the interaction or contact zone. Also, *F. arthrosporioides*, *F. flocciferum*, *F. poae*, and *F. torulosum* did not appear to be suitable hosts for *S. mycoparasitica*. Around five days after inoculation



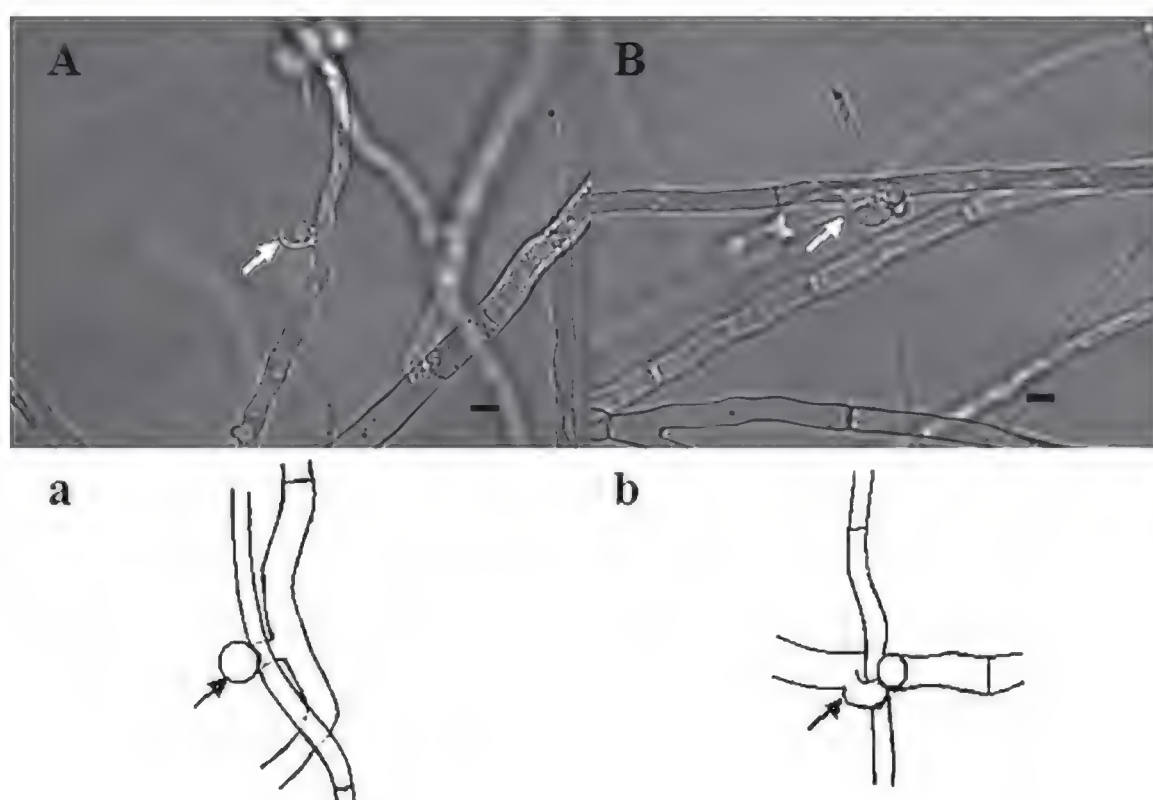


FIG. 1. *Sphaerodes mycoparasitica*-*Fusarium* spp. mycoparasitism assays. (A-a). Hook-shaped contact structures (arrows). (B-b). Clamp-like clasping cells (arrows). Figures a and b are diagrammatic drawings for both A and B. Scale bars = 5 μm.

on slide culture assays, mycelia of *F. arthrosporioides* were inhibited by *S. mycoparasitica*. *Fusarium arthrosporioides* started to form rosette-like mycelia at the contact zone with *S. mycoparasitica* (FIG. 2B).

On the fifth and seventh days after inoculation, anamorphic structures were produced by *S. mycoparasitica* more abundantly in the zone of contact with *F. culmorum* (FIG. 2C, D). Anamorphic structures or asexual organs in close proximity to *F. culmorum* mycelia were red-colored (FIG. 2D), whereas the organs at a distance were not (FIG. 2C).

### Fungus-fungus coevolution

Six *Sphaerodes* and one *Melanospora* species — *S. compressa* (Udagawa & Cain) P.F. Cannon & D. Hawksw., *S. fimicola* (E.C. Hansen) P.F. Cannon & D. Hawksw., *S. mycoparasitica*, *S. quadrangularis*, *S. retispora* var. *retispora*, *S. singaporensis* (Morinaga, Minoura & Udagawa) Dania García, Stchigel & Guarro, *Melanospora brevirostris* — were phylogenetically analysed. Information related to these strains is summarized in TABLE 1. Node M<sub>1</sub> is the point of divergence between the three *Fusarium*-specific *Sphaerodes* spp. and the other four taxa (FIG. 5; TABLE 1).

The phylogenetic tree further shows that the three *Sphaerodes* mycoparasites of *Fusarium* species — *S. mycoparasitica*, *S. quadrangularis* and *S. retispora* —

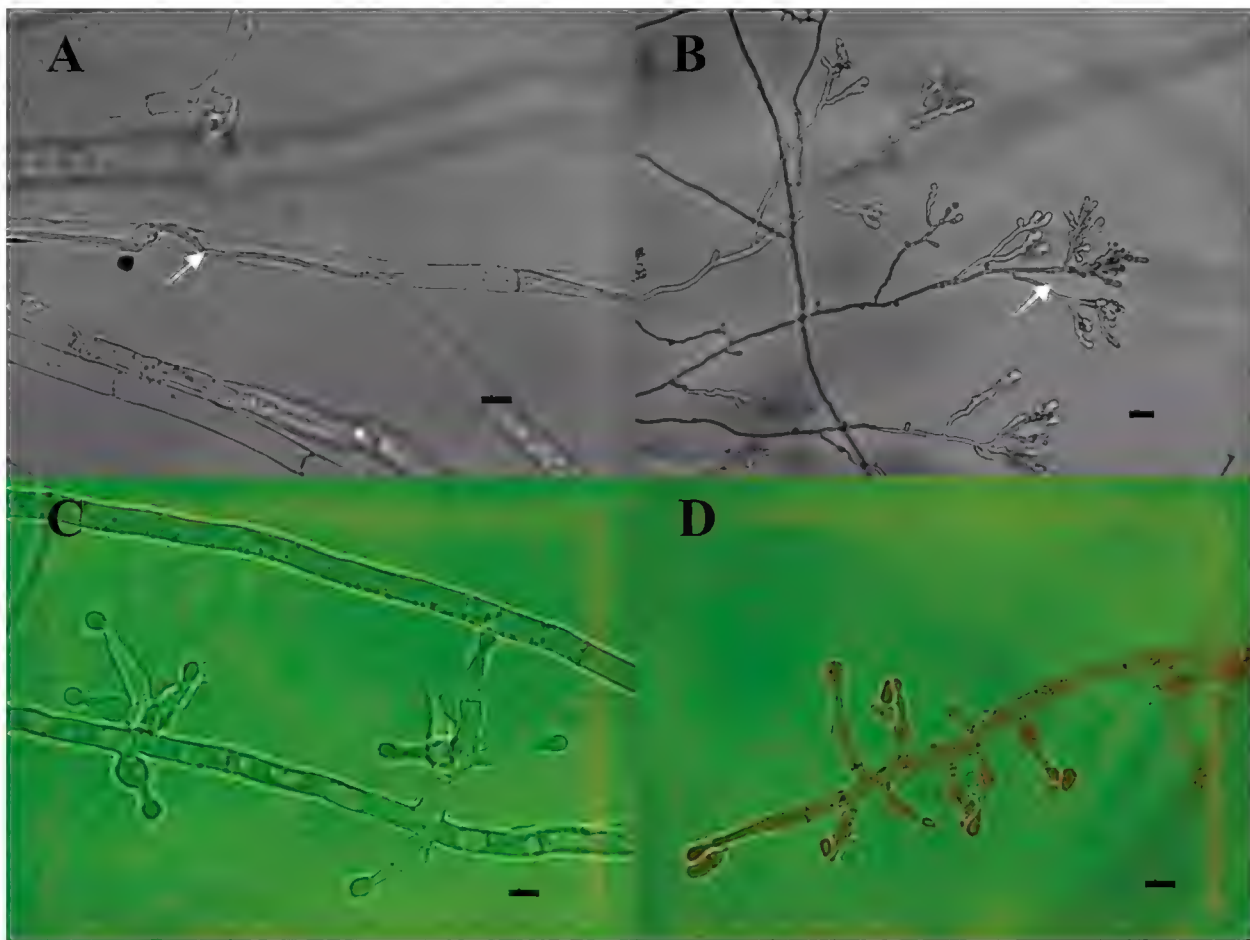


FIG. 2. Intracellular parasitism, hyphal inhibition response, and anamorphic stages during the *Sphaerodes mycoparasitica*–*Fusarium* spp. interactions. (A). Intracellular parasitism by *S. mycoparasitica* in *F. equiseti* (arrow). (B). *Fusarium* hyphal inhibition response when challenged with *S. mycoparasitica*; deformation of hyphae into rosette-like shapes (arrow). (C). Hyaline *S. mycoparasitica* anamorphic stages. (D). *Sphaerodes mycoparasitica* anamorphic stages with adsorption of red pigments from *F. culmorum*. Scale bars A, C, D = 5µm; B = 20µm.

diverge at  $M_2$  to distinguish hyperparasites on white-pigmented *F. oxysporum* (such as *S. retispora*) from those on a red-pigmented *F. avenaceum* host (such as *S. quadrangularis*). Moreover, evolution from  $M_2$  occurs at  $M_3$  giving rise to mycoparasites of white- and red-pigmented *Fusarium*. This is the case of *S. mycoparasitica*, which attacks *F. avenaceum*, *F. culmorum*, *F. equiseti*, *F. graminearum*, and *F. oxysporum*, (FIG. 5; TABLE 1). Thus,  $M_3$  is the point where polyspecificity as opposed to monospecificity on *Fusarium* appears.

## Discussion

The small knobs or hook-shaped contact structures formed by *Sphaerodes mycoparasitica* on *Fusarium culmorum* and *F. equiseti* were similar to those described by Whaley & Barnett (1963) for *Gonatobotrys simplex* Corda [= *Melanospora damnosa* (Sacc.) Lindau] on *Alternaria tenuis* Nees [*A. alternata*], and by Jordan & Barnett (1978) for *Melanospora zamiae* Corda on *Tritirachium*

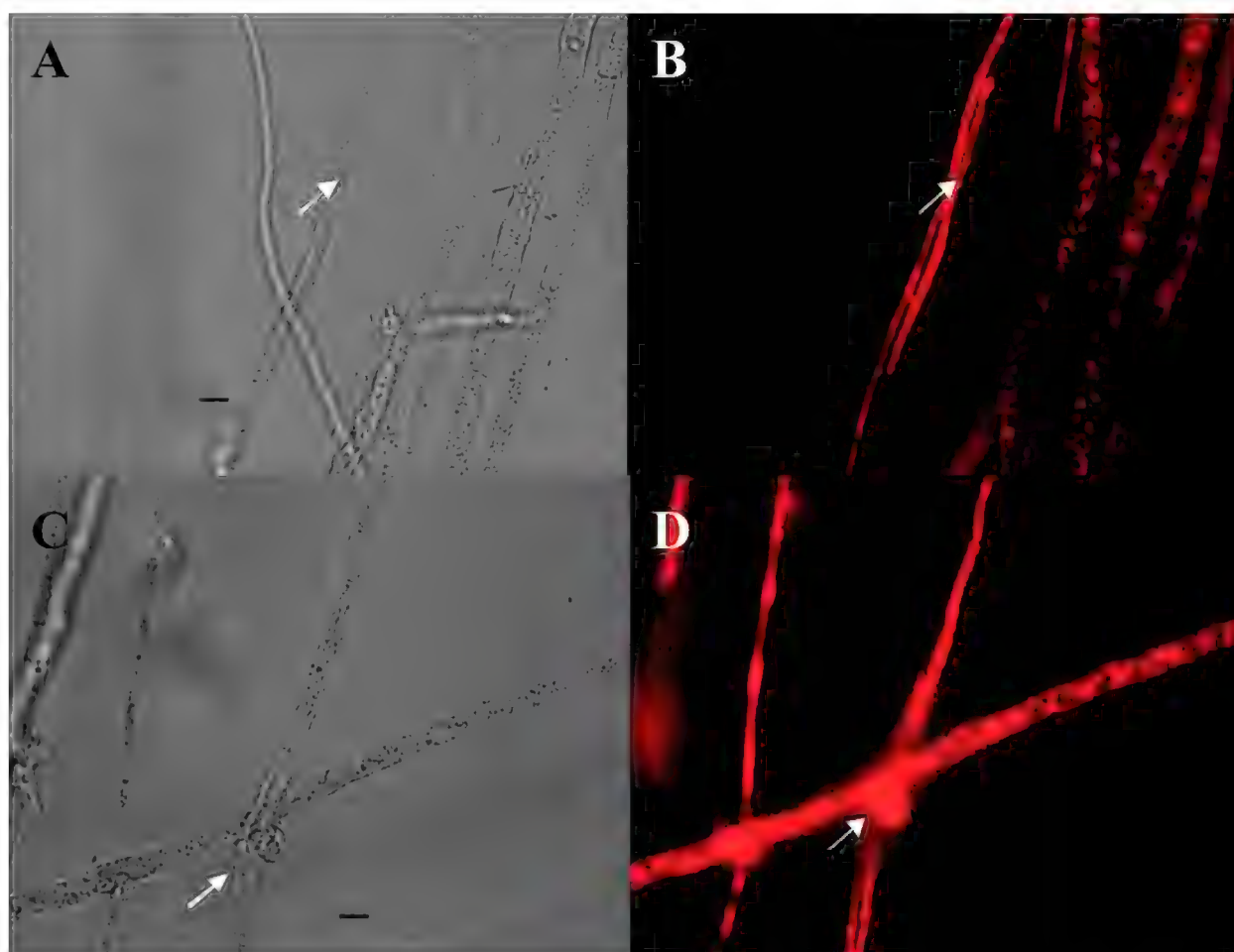


FIG. 3. (A–B) Intracellular parasitism by *Sphaerodes* inside *Fusarium equiseti* (arrows). (B–D) Intracellular hyphae produced by *Sphaerodes* inside *F. equiseti* with hook-shaped contact structure (arrows). A and C were captured under light microscopy; whereas in B and D hyphae were stained with lactofuchsin and images were captured under fluorescent and confocal laser microscopy, respectively. Scale bars = 5µm.

sp. Hook-shaped contact structures are well-known among biotrophic mycoparasites in the *Melanosporales*. Harveson & Kimbrough (2001) were the first to report *S. retispora* var. *retispora* as a contact biotrophic mycoparasite on *F. oxysporum* with hook-like contact structures. Harveson & Kimbrough (2001) also reported another melanosporaceous fungus, *Persiciospora moreaui* P.F. Cannon & D. Hawksw., as a contact biotrophic mycoparasite of *F. oxysporum* with similar contact branches as in *M. zamiae* and *S. retispora* (Harveson & Kimbrough 2000). Recently, *S. mycoparasitica* was found to produce similar hook-shaped contact structures on *Fusarium avenaceum*, *F. graminearum*, and *F. oxysporum* (Vujanovic & Goh 2009) and *S. quadrangularis* on *F. avenaceum* species (Goh & Vujanovic 2010). In this study, *S. mycoparasitica* was observed to form clamp- or clasp-like contact branches to attach to *F. equiseti* and *F. culmorum* (FIG. 1B, b). These structures were also reported for *Stephanoma phaeosporum* E.E. Butler & McCain, another biotrophic mycoparasite



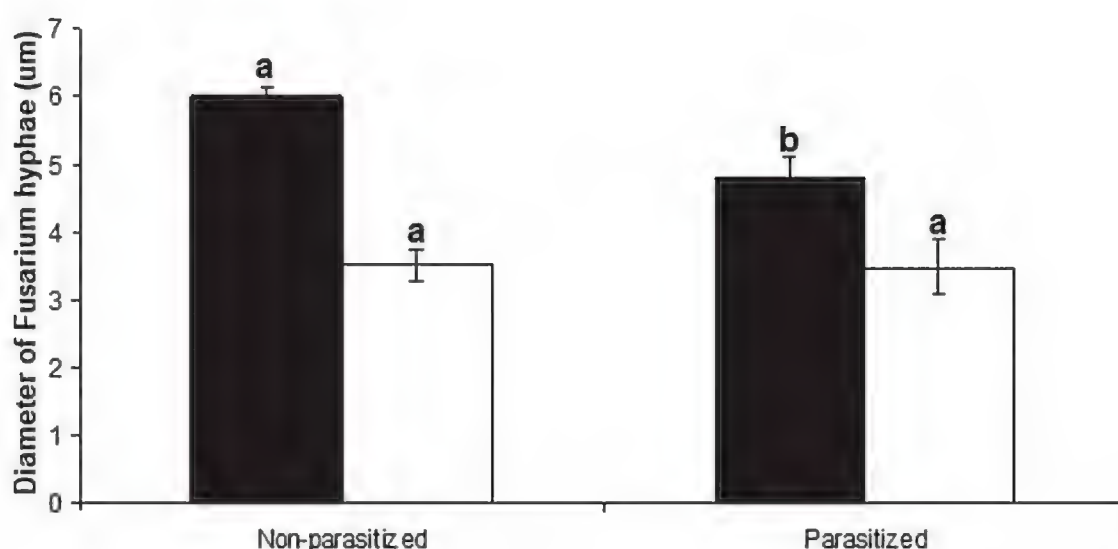


FIG. 4. Mean hyphal diameters of parasitized and non-parasitized *Fusarium equiseti* cells (■) and *F. culmorum* (□) on 1-week slide-cultures with *Sphaerodes mycoparasitica* biotrophic mycoparasite. Data are means and standard deviations. Same lowercase letters indicate no significant difference between parasitized and non-parasitized hyphae at  $P = 0.05$ , with T-test.

(Rakvidhyasastra & Butler 1973). These contact structures may also be employed by contact or fusion biotrophic mycoparasites as tools to acquire nutrients from the hosts (Carmichael 1955; Gams et al. 2004; Whaley & Barnett 1963). Nutrients, growth factors, biotins, mycotrophein, and thiamine have been found to be important for nourishment and proliferation of biotrophic mycoparasites (Hwang et al. 1985; Jordan & Barnett 1978).

In this study, loss of cytoplasm (FIG. 3A, C) and a reduction of the diameter of *F. equiseti* hyphae resulted from mycoparasitism (FIG. 4). Similarly, Harveson & Kimbrough (2001) noticed that *Sphaerodes retispora* and *M. zamiae* isolates reduced the total hyphal weight and aerial hyphae of *F. oxysporum*, in addition to inhibiting the growth of this *Fusarium* species. Furthermore, loss or decreased intensity of staining or colour of dye in host cells (compared to healthy) were further reported by White & Traquair (2006) as an indication of loss of cytoplasm and intracellular infection. Intracellular parasitic activity was also described in *Fusarium*–*Rhizoctonia* and *Mucor*–*Rhizopus* mycoparasitic interactions (Arora & Dwivedi 1980; Gupta & Tandon 1978; Gupta et al. 1979). Although hyphal diameter of *F. culmorum* was not reduced by *S. mycoparasitica* (FIG. 4), this could be due to the lack of intracellular penetration in *F. culmorum* during the tested period. Barnett (1963), Jordan & Barnett (1978), Jeffries & Young (1994) and Jeffries (1995), have all pointed out that biotrophic mycoparasites, in general, have narrow host ranges. Therefore, it is not surprising that not all the *Fusarium* taxa tested could act as hosts for *S. mycoparasitica*, *S. quadrangularis* and *S. retispora*.

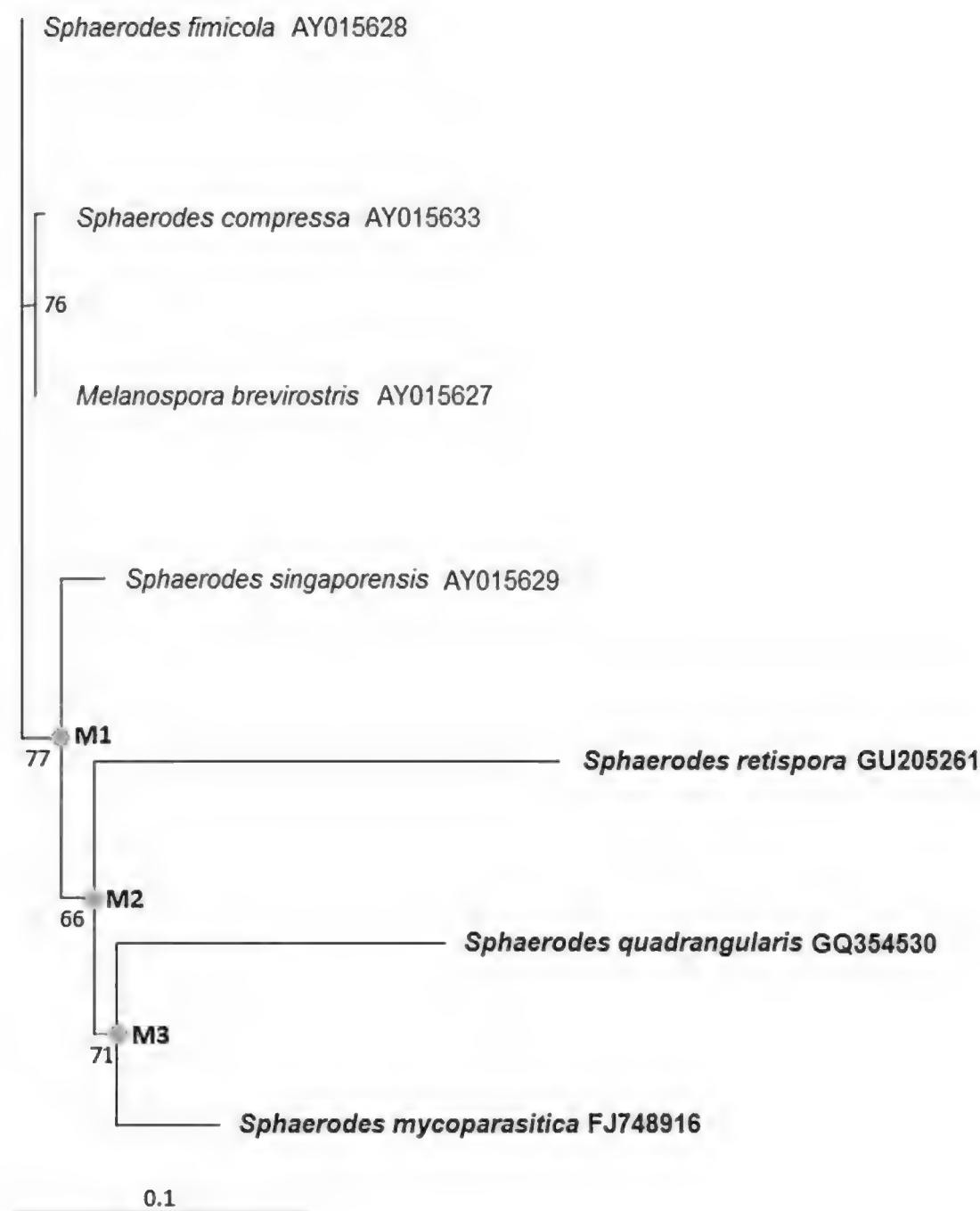


FIG. 5. Phylogenetic tree based on LSU rDNA sequences for six *Sphaerodes* species showing position of mycoparasitic taxa associated with *Fusarium* hosts. M1 – the point of evolutionary divergence between *Sphaerodes* mycoparasites associated with *Fusarium* and *Sphaerodes* taxa including closely related *Melanospora brevirostris* associated with other fungal and plant hosts; M<sub>2</sub> – point of branching towards specialization or monospecificity of *S. retispora* on *F. oxysporum* (white mycelium) and monospecificity of *S. quadrangularis* on *F. avenaceum* (red mycelium); M<sub>3</sub> – the point of evolutionary direction towards polyspecificity of *S. mycoparasitica* on various white and red *Fusarium* hosts. Bootstrap values of 50% or greater from 1000 bootstrap replications are indicated for the corresponding branches.



TABLE 1. Information related to six *Sphaerodes* species and a *Melanospora* species used for phylogenetic analysis.

	ISOLATED FROM	MYCO- PARASITE	DISTRIBUTION	REFERENCE
<i>Melanospora brevisporis</i> *	Dead plant stems and decaying truffles as well as on various <i>Pezizales</i> , usually <i>Sepultaria</i> sp.	Yes	England, North Europe	Cannon et al. 1985; Cannon & Hawksworth 1982; Farr & Rossman 2009
<i>Sphaerodes compressa</i>	Soil, cow dung, dead leaves, aerial contaminant	No	Canada, USA, Japan, New Caledonia	Cannon et al. 1985; Farr & Rossman 2009
<i>S. fimicola</i>	Dung, surface litter and soil, plants	No	Europe, USA, Madeira, British Isles	Cannon et al. 1985; Farr & Rossman 2009
<i>S. mycoparasitica</i>	Several <i>Fusarium</i> species	Yes	Canada	Vujanovic & Goh 2009
<i>S. quadrangularis</i>	<i>F. avenaceum</i>	Yes	Spain	García et al. 2004; Goh & Vujanovic 2010
<i>S. retispora</i> var. <i>retispora</i>	<i>F. oxysporum</i>	Yes	Japan, New Guinea, USA	Cannon et al. 1985; Harveson & Kimbrough 2001
<i>S. singaporensis</i>	Soil	Unknown	Singapore	Morinaga et al. 1978

\*Note: Information on *Melanospora brevisporis* (Fuckel) Höhn. was also included since the LSU rDNA sequences analyses in Fig 5. of this article suggest relatedness to *S. compressa* and *S. fimicola* in concordance with findings of Davey et al. (2008).

*Sphaerodes quadrangularis* was first described by García et al. (2004). At the time its anamorph was unknown. Here, *S. quadrangularis* was observed to produce mono- and polyphialides or asexual organs like those of *S. mycoparasitica* (FIG. 2C) and *S. retispora* (Harveson & Kimbrough 2001) when inoculated together with *Fusarium avenaceum*. Based on *S. mycoparasitica* analyses, Vujanovic & Goh (2009) proposed that most anamorphic traits in *Sphaerodes* (e.g., hyaline and ampulliform phialides as well as irregularly branched conidiophores) resemble those of *Trichoderma* species (sect. *Pachybasium*) in *Hypocreales*. In contrast, the base-to-neck ratios of phialides in *S. mycoparasitica*, *S. quadrangularis*, and *S. retispora* show interspecies differences summarized in the key to taxa of *Sphaerodes*.

**Key to the mycoparasitic taxa of *Sphaerodes***

- 1 Clamp-like contact structures present .....2
- 1\* Clamp-like contact structures lacking. Phialides with base-neck width ratio < 2; mono- and polyphialidic anamorphic stages; monospecific on *F. oxysporum* ..... *S. retispora* var. *retispora*
- 2 Intracellular penetration and haustoria present. Phialides with base-neck width ratio between 2–2.5; mono- and polyphialidic anamorphic stages; polyspecific on *F. avenaceum*, *F. culmorum*, *F. equiseti*, *F. graminearum*, and *F. oxysporum* ..... *S. mycoparasitica*
- 2\* Intracellular penetration and haustoria absent. Phialides with base-neck width ratio > 2.5; mono- and polyphialidic anamorphic stages; monospecific on *F. avenaceum* ..... *S. quadrangularis*

In addition, this study showed that when *S. mycoparasitica* and *F. culmorum* were co-inoculated in slide culture, anamorphic structures and hyphae of the former were red-colored (FIG. 2D). Similarly, *S. quadrangularis* hyaline hyphae became red-colored after contacting *F. avenaceum* hyphae (Goh & Vujanovic 2010). This could be due to the absorption of *Fusarium* red pigments by *Sphaerodes* through biotrophic mycoparasitism (Goh & Vujanovic 2010). However, the mechanism of this phenomenon remains unclear. The red pigments of *F. avenaceum*, *F. culmorum*, and *F. graminearum* are aurofusarin toxins (Malz et al. 2005). Perhaps host toxins drive the evolution of mycoparasites. Thus, it would be interesting for further studies, as indicated by relatedness of these *Fusarium*-specific *Sphaerodes* taxa (FIG 5.), to explore whether it is actually the nature of fusaria toxins that create an evolutionary pressure inducing specialization within *Sphaerodes*.

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# MYCOTAXON

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## **Additional and new lichen records from Cozia National Park, Romania**

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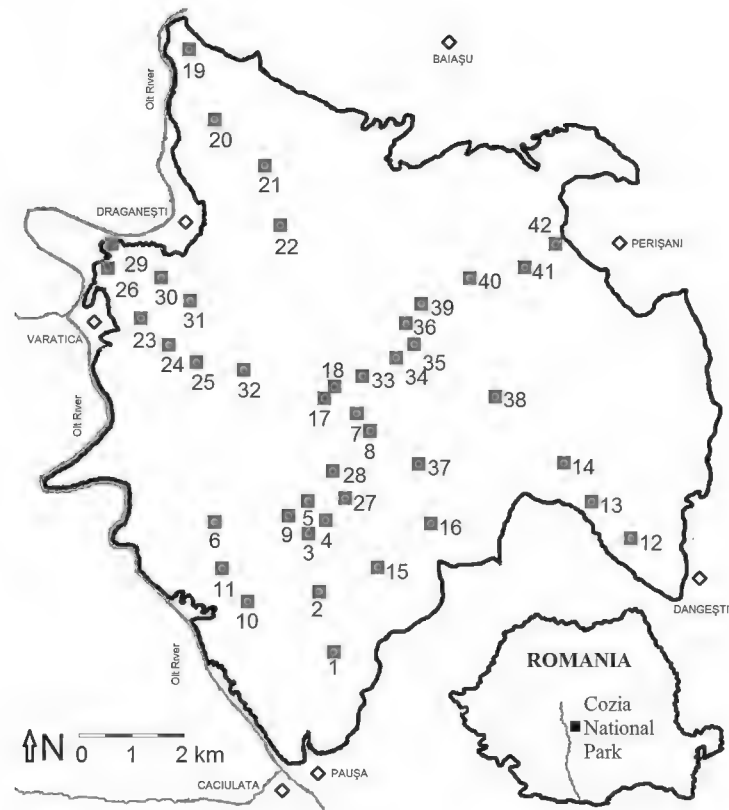
**Abstract** — A list of 115 lichen taxa from Cozia National Park includes 8 new records for the mycota of Romania and 77 taxa new for Cozia. Distribution and substrata are summarized, and the complete annotated species list is posted at <http://www.mycotaxon.com/resources/weblists.html>

**Keywords** — lichenized fungi, biodiversity, biota, checklist, Cozia Mount

## **Introduction**

The present study of the lichen diversity on Cozia Mount, the primary massive area of Cozia National Park, aims to contribute to the lichen biota of Romania. As one of the most detailed lichenological surveys in recent years, the report lists 115 taxa, of which 77 are new for Cozia National Park and Valcea County and 8 are new to Romania.

Romanian lichens have been studied for over 150 years and the reports are cited in over 300 publications, including a survey of all available mycological information by Moruzi et al. (1967). Ciurchea (1998, 2007a,b) subsequently



Map of the study area — Cozia Mount and surrounding villages with sampling site numbers.

revised comprehensively the checklist of lichens and lichenicolous fungi for Romania, now available online (<http://www.bgbm.org/BGBM/STAFF/Wiss/Sipman/Zschackia/Rumania/index.htm>).

The lichens of Cozia Mount have previously been studied by Codoreanu & Ciurchea (1965), Ciurchea (1969, 1970), Bartók (1990), Costache et al. (2007), and Çobanoğlu et al. (2009).

### Materials and methods

Cozia National Park is situated on the central-southern region of Romania, in Valcea County, inside the southern Carpathians. Cozia Mount (Ciuha Neamtului) is the highest peak, with its 1668 meter summit. It is intersected from north to south by the Olt River (FIG. 1). The climate is specific to mountain depressions without large temperature variations, with cool summers (about 20°C in July), relatively mild winters (between –5 and 0°C in January), and an average annual temperature of 9°C. Precipitation is moderate, 750–800 mm annually (Ploaie 2004).

Lichens were collected from 42 different sites on Cozia Mount, located on the East side of Olt River in Cozia National Park, Valcea County. Specimens were investigated microscopically (Olympus SZx40) and chemically by using spot tests (standard K, C, P and I) following Purvis et al. (1992). The taxa were identified to the level of species (except two genera) with the aid

of identification keys (Brodo et al. 2001, Purvis et al. 1992, Wirth 1995). The collections are preserved in the Herbarium of the Faculty of Science and Arts, Marmara University, Istanbul (MUFE), and duplicates have been stored in the Herbarium of the University of Craiova (Romania).

## Results

The list of identified lichens cites 115 taxa in 61 genera in alphabetical order. Nomenclature mainly follows Index Fungorum ([www.indexfungorum.com](http://www.indexfungorum.com)) and the recent literature (Ahti & Hawksworth 2005, Blanco et al. 2004). Author names are abbreviated according to Brummitt & Powell (1992). Eight taxa are new to Romanian lichen mycota, and 77 taxa are newly recorded from Cozia Mount. Also 26 taxa are rare for Romanian mycota according to Ciurchea (2007a,b).

## Discussion

Among the 115 taxa recorded, the eight recorded as new to Romania include *Buellia griseovirens*, *Candelariella coralliza*, *Cladonia stellaris*, *Lecanora cinereofusca*, *Leproloma cacuminum*, *Ochrolechia inaequatula*, *Trapelia involuta*, and *Usnea silesiaca*. Seventy-seven taxa are new to Cozia Mount. Additionally, among the 26 species regarded as rare in Romania (Ciurchea 2007a,b) are *Cornicularia normoerica*, *Immersaria athroocarpa*, *Lecidella carpathica*, *Melanelia stygia*, *Ophioparma ventosa*, *Protoblastenia incrustans*, and *Sphaerophorus fragilis*. The majority of the lichen taxa designated in the list is saxicolous (89 taxa, or 77% of the total). Of the saxicolous lichens, siliceous taxa (51) are dominant followed by calcareous taxa (27), and those reported on sandstone (11). Morphologically, 81 taxa are crustose (70.4%), 20 foliose (17.4%), 9 fruticose (7.8%), one squamulose (0.9%) and four dimorphic *Cladonia* spp. (3.5%).

The present study, which represents the most detailed recent lichenological survey in Romania, provides valuable data for the lichen mycota.

## Acknowledgements

This study is a part of research project supported by The Research Fund of Marmara University with the Project Number FEN-BGS-120707-0154. Gülşah Çobanoğlu was funded at the beginning of the project by the scholarship of Türk Petrol Vakfı, and Mustafa Yavuz was funded by TUBITAK (Turkish Scientific and Technical Research Council (TUBITAK BİDEB 2214-2007)).

We are thankful to Ex. Grand Ambassador of Turkey in Bucharest, his Excellency Mr. Ahmet Rıfat Ökçün; special thanks to Dr. Daniel Radutoiu from University of Craiova; the Manager of the Cozia National Park, Mr. Pavel Prundurel; the Ranger Mr. Narcis Olteanu; Ms. Birkan Açıkgoz (MsD), and Mr. Laurentiu Baloniu (MsD) for assistance

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## MYCOTAXON

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**A new *Asterostroma* species (*Basidiomycota*)  
from a subtropical region in Japan**HIROTO SUHARA<sup>1\*</sup>, NITARO MAEKAWA<sup>1</sup> & SHUJI USHIJIMA<sup>2</sup>*h\_suhara@muses.tottori-u.ac.jp & kin-maek@muses.tottori-u.ac.jp*<sup>1</sup>*Faculty of Agriculture, Tottori University**4-101 Koyama-Minami, Tottori, 680-8553, Japan**ushi-kintai@go4.enjoy.ne.jp**The United Graduate School of Agricultural Science, Tottori University**4-101 Koyama-Minami, Tottori, 680-8553, Japan*

**Abstract** — A new homobasidiomycete, *Asterostroma boninense*, was found in the Bonin (Ogasawara) Islands, a subtropical region in Japan. This species is morphologically characterized by having resupinate basidiomata, a monomitic (asterodimitic) hyphal system, simple septate generative hyphae, dextrinoid asteroetae, four sterigmata basidia, and subglobose, tuberculate, amyloid basidiospores. It is similar to *A. muscicola*, but the latter has smaller basidia. In Japan, *A. muscicola* is widely distributed in warm-temperate to subtropical regions including the Bonin Islands, while *A. boninense* is restricted to the Bonin Islands. Another species in the genus, *Asterostroma andinum*, is also reported as new to Japan. A key to the Japanese species of *Asterostroma* is provided.

**Key words** — corticioid fungi, *Lachnocladiaceae*, oceanic island, taxonomy

**Introduction**

The genus *Asterostroma* Masee belonging to the family *Lachnocladiaceae* (*Basidiomycota*) is characterized by resupinate and felted-membranous basidiomata, gloeocystidia, clampless generative hyphae, and dextrinoid asteroetae (asterohyphidia). Based on basidiospore morphology, the genus is divided into two subgenera, *Austroasterostroma* Parmasto and *Asterostroma*. The former produces smooth and inamyloid basidiospores whereas species of the latter have amyloid spores (Parmasto 1970). Furthermore, the subgenus *Asterostroma* is subdivided into two sections, *Laevispora* Parmasto (with smooth basidiospores) and *Asterostroma* (with ornamented basidiospores) (Parmasto 1970, Boidin et al. 1997). According to MycoBank administered by the International Mycological Association (<http://www.mycobank.org/>),

**NOTE** — MYCOTAXON prepared this PDF with color plates for the author. The original print version was published with halftone (grayscale) plates.



twenty-six species have been described in *Asterostroma*. Among them, *A. cervicolor* (Berk. & M.A. Curtis) Massee (Aoshima et al. 1963), *A. macrosporum* N. Maek. & Suhara (Suhara et al. 2010), and *A. muscicola* (Berk. & M.A. Curtis) Massee (Suhara et al. 2010) have been earlier reported from Japan. In the present study, we describe a new species of the genus based on specimens collected in the Bonin (Ogasawara) Islands, located about 1000 km south of Tokyo, Japan. Moreover, an additional species of *Asterostroma* is reported as new to Japan.

### Materials & methods

The specimens are deposited in the Tottori University Fungal Herbarium (TUFH) and the cultures in the Tottori University Mycological Culture Collection (TUMC). Morphological observations were carried out as described in Suhara et al. (2010). Color names in double quotation marks are based on Rayner (1970). The notation “basidiospores (n = 60/3)” indicates that measurements were made on 60 spores from 3 specimens. Polysporous isolates obtained from each specimen were grown on malt extract agar [MA; 1.5% (w/v) malt extract and 1.5% (w/v) bacto agar, Difco, Detroit, MI, USA]. To determine the optimum growth temperature, the isolates were grown on MA plates at 8 different temperatures: 4, 10, 15, 20, 25, 30, 35 and 40°C.

### Taxonomy

*Asterostroma boninense* Suhara & N. Maek., sp. nov.

FIGS. 1–7

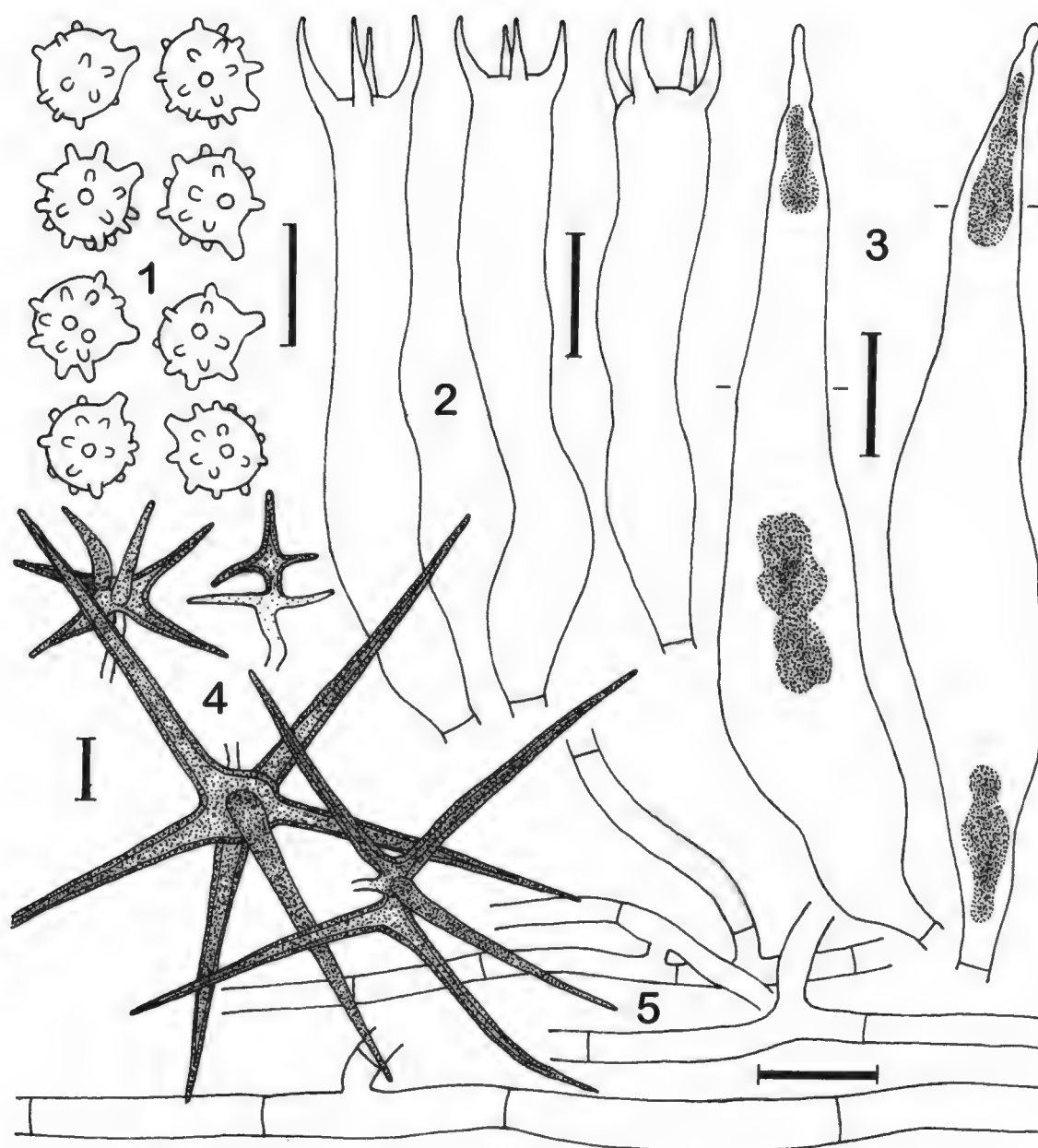
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*Basidiomata resupinata, adnata, effusa, mollia, 200–600 µm crassa; superficies hymenialis “Buff” vel “Ochreous” (sec. Rayner 1970), laevis, sub lente (×20); margo “Ochreous”, “Fulvous” vel “Cinnamon”, tenuescens, interdum fimbriatus, filis hyphalibus tenuibus nonnumquam. Systema hyphale monomiticum; hyphae generatoriae cum septis, sine fibulis, 1.5–5 µm diametro, laeves, tenui-vel parum crassitunicatae (usque 0.5 µm), simpliciseptatae; asterohyphidia numerosa, radii ad 110 µm longi. Cystidia (gloeocystidia) parum numerosa, subcylindracea, ventricosa vel fusioidea, 43–95 × 7.5–16 µm. Basidia subcylindrica vel utriformia, 40–60 × 6.5–8.5 µm, 4 sterigmata gignentia. Basidiosporae subglobosae apiculo distincto armatae, 5.5–7.5 × 5–7.2 µm (praeter tuberculis), tubercula (tuberculis usque ad 1.5 µm longi), tenuitunicatae, amyloideae.*

TYPE: JAPAN. TOKYO: Ogasawara-mura, Takinoura (Anijima Island), on dead trunk of *Clinostigma savoryanum* (Rehder & E.H. Wilson) H.E. Moore & Fosberg (*Arecaceae*), 6 Dec 1997, coll. N. Maekawa. (Holotype, TMI20619; ex-type culture (polysporous), TUF33876).

ETYMOLOGY: The specific epithet *boninense* refers to the geographic origin of the type specimen.

Basidiomata resupinate, loosely adnate, effused, soft, felt-like, 200–600 µm thick; hymenial surface “Buff”, partly “Ochreous”, smooth, pruinose under



FIGS 1–5. Line drawings of *Asterostroma boninense* (TMI20619, holotype): 1. Basidiospores; 2. Basidia; 3. Cystidia (gloeocystidia) – short horizontal lines indicate the level of the hymenial surface; 4. Asterohyphidia (asterosetae); 5. Subicular hyphae. Scale bar = 10  $\mu$ m.

the lens ( $\times 20$ ), sometimes slightly cracked when dried; margin “Ochreous”, “Fulvous” to “Cinnamon”, determinate, but sometimes thinning out, fimbriate, occasionally with thin hyphal strands concolorous with the margin under the lens ( $\times 20$ ). Context in vertical section ocher, pellicular to submembranous, the subiculum sometimes with thin hyphal strands and/or containing crystals. Hyphal system monomitic (asterodimitic); generative hyphae 1.5–5  $\mu$ m in diameter, smooth, thin- to slightly thick-walled (up to 0.5  $\mu$ m), clampless-septate, loosely intertwined in the subiculum; asterohyphidia (asterosetae)

numerous in the subiculum and subhymenium, subhyaline to brownish, 2–10 diverging branches, the branches acicular to subulate, up to 110  $\mu\text{m}$  in length; cystidia (gloeocystidia) subcylindrical, ventricose to fusiform, sometimes with schizopapillae, 43–95  $\times$  7.5–16  $\mu\text{m}$ , without a basal clamp, thin-walled, with pale yellowish oily contents, imbedded in the basidiomata, but sometimes projecting 30  $\mu\text{m}$  beyond the hymenial surface; basidia ( $n = 60/3$ ) subcylindrical to utriform, 40–60  $\times$  6.5–8.5  $\mu\text{m}$ , thin-walled, without a basal clamp, consistently producing 4 sterigmata; basidiospores ( $n = 60/3$ ) subglobose, 5.5–7.5  $\times$  5–7.2  $\mu\text{m}$  (excluding tubercles), with a distinct apiculus, tuberculate (tubercles up to 1.5  $\mu\text{m}$  in length), thin-walled, amyloid.

DISTRIBUTION — So far only reported from the Bonin Islands (Japan).

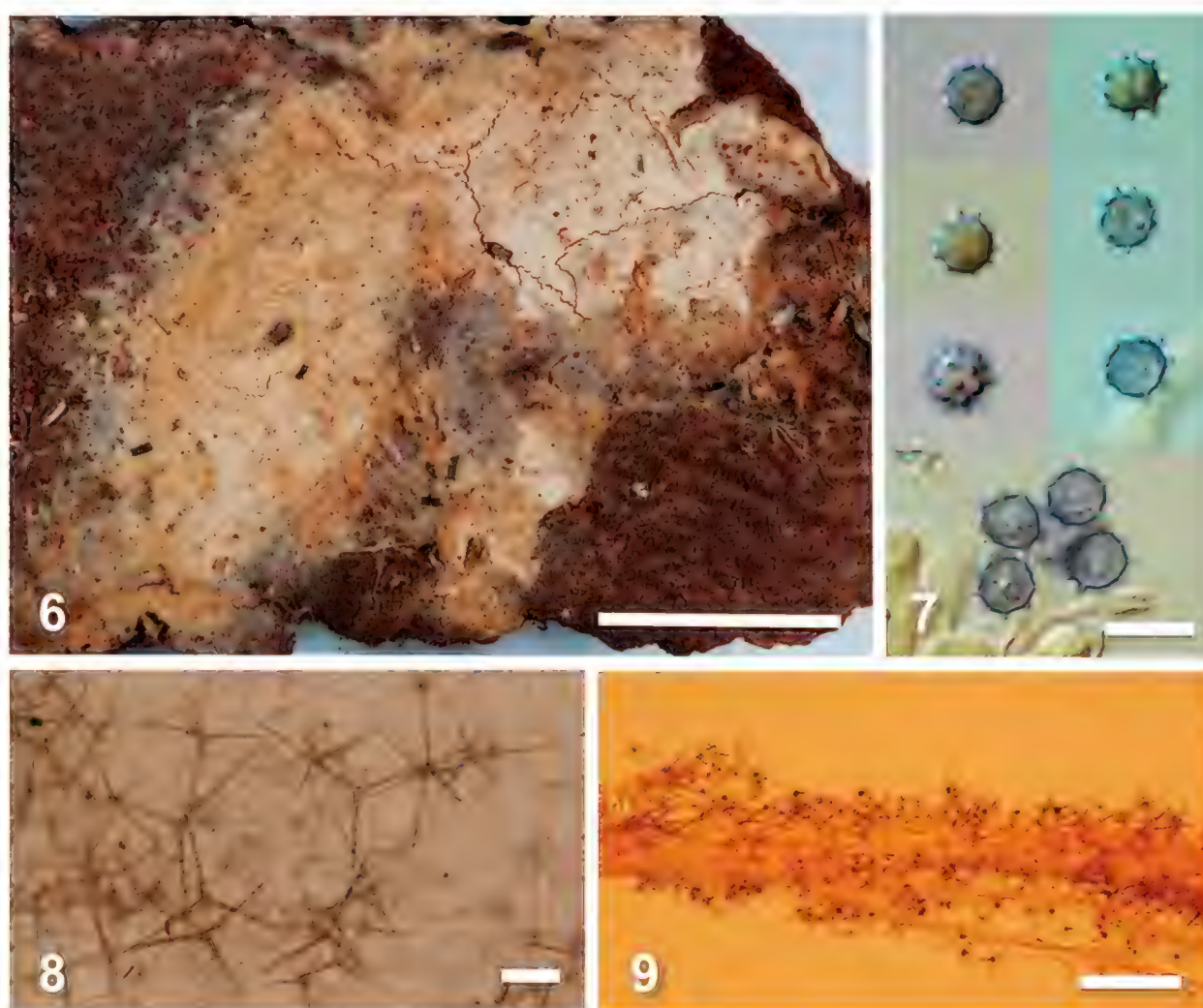
CULTURAL CHARACTERISTICS — Optimal temperature for the four polysporous isolates examined was 25–30°C (see TYPE and ADDITIONAL SPECIMENS EXAMINED). These isolates grew between 10 and 30°C, with no visible growth observed at 4, 35, or 40°C. Growth rate on MA: 6.7–15 mm after 1 w (25°C).

ADDITIONAL SPECIMENS EXAMINED: JAPAN. TOKYO: Ogasawara-mura, MINAMIZAKI (Hahajima Island), on dead wood of *Livistona boninense* (Becc.) Nakai (*Arecaceae*), 12 Dec 1997, coll. N. Maekawa, TMI20570 (polysporous culture, TUFC33791); TAKINOURA (Anijima Island), on dead trunk of *L. boninensis*, 6 Dec 1997, coll. N. Maekawa, TMI20620 (polysporous culture, TUFC33877); MT. SHIGURE (Chichijima Island), on dead branch of *Pandanus boninensis* Warb. (*Pandanaceae*), 3 Dec 2006, coll. N. Maekawa, TUMH40170 (polysporous culture, TUFC10922).

Mycelial mats white, partly pale salmon to “Flesh”, cottony to woolly at 2 w, and then becoming partly floccose, “Rosy Vinaceous” to “Dark Vinaceous”, sometimes with white, thin, hyphal strands, occasionally farinaceous around the inoculum; agar medium stained “Vinaceous” around the inoculum at 6 w; margin even, raised, with irregularly fan-like extensions; odor crayon-like; no fruiting by 6 w. Surface and aerial hyphae hyaline, 2.5–3.5  $\mu\text{m}$  in diameter, smooth, thin-walled, clampless-septate, sparsely branched, sometimes with yellow to reddish brown oily contents, producing abundant subhyaline to pale brown asterohyphidia (FIG. 8), occasionally producing tubular gloeocystidium-like cells and gloeoplerous to swollen (monilioid) cells, up to 20  $\mu\text{m}$  in diameter filled with hyaline oily contents. Hyphae of the hyphal strands hyaline to subhyaline, 1–3  $\mu\text{m}$  in diameter, smooth, thin-walled, clampless-septate, sparsely branched, producing numerous subhyaline to pale brown asterohyphidia (FIG. 9), sometimes containing crystals in hyphal strands. Submerged hyphae hyaline to subhyaline, partly becoming pale “Vinaceous”, 1–2.5  $\mu\text{m}$  in diameter, smooth, thin-walled, clampless-septate, branched, sometimes capilliform-like; skeletal and binding hyphae absent.

Species code (Nakasone 1990): 6. 15. 16. 19. 26. 28. 29. (31.) 36. 39. 44. 49. 53. 54.





FIGS 6–9. Photographs of *Asterostroma boninense*: 6. Basidioma (TMI20619, holotype); 7. Basidiospores stained with Melzer's reagent; 8. Asterohyphidia produced in cultural mycelium (6 w); 9. Hyphal strand with asterohyphidia produced in culture (6 w). Scale bars: 6 = 1 cm; 7, 8 = 10  $\mu\text{m}$ ; 9 = 100  $\mu\text{m}$ .

## Discussion

*Asterostroma boninense* is primarily characterized by having asterohyphidia and tuberculate, subglobose, amyloid basidiospores. Its amyloid and ornamented basidiospores places this species into subg. *Asterostroma* sect. *Asterostroma*. Within this section, the species resembles *A. muscicola* and *A. macrosporum* in forming subglobose basidiospores with subcylindrical to obtuse ornaments. However, Gilbertson & Blackwell (1987) and Boidin et al. (1997) measured the basidia of *A. muscicola* at  $25\text{--}32 \times 6\text{--}8.5 \mu\text{m}$  and  $18\text{--}24 \times 5\text{--}6 \mu\text{m}$  respectively; the distinctly larger basidia in *A. boninense* [ $40\text{--}60 \times 6.5\text{--}8.5 \mu\text{m}$  ( $50 \pm 7.4 \times 7.4 \pm 0.5 \mu\text{m}$ ,  $n = 60/3$ )] differentiate the new species from *A. muscicola* [ $27\text{--}41 \times 5\text{--}7.5 \mu\text{m}$  ( $34.2 \pm 3.8 \times 6.3 \pm 0.7 \mu\text{m}$ ,  $n = 40/2$ )]. Furthermore, *A. boninense* specimens have been collected only from dead monocotyledonous angiosperm tree trunks and branches, e.g., endemic species of *Clinostigma*, *Livistona*, and *Pandanus* in the Bonin Islands (located in subtropical region of Japan). On the

other hand, *A. muscicola* occurs both on angiospermous and gymnospermous slash (Gilbertson et al. 1974, Gilbertson & Blackwell 1987) and is distributed in subtropical to warm-temperate regions in Japan (Suhara et al. 2010).

*Asterostroma boninense* also resembles *A. macrosporum* in basidial shape and size except that in the latter basidiospores are distinctly larger ( $8.5\text{--}11 \times 7.5\text{--}9\text{ }\mu\text{m}$ ) than those of *A. boninense*. In addition, *A. macrosporum* has been collected only from mangrove trees on Iriomote Island, approximately 1,600 km west of the Bonin Islands (Suhara et al. 2010).

We also recognized *A. andinum* Pat. as a species new to Japan based on two specimens, TMI19638 and TUMH40171, collected in Hokkaido and the Bonin Islands, respectively. This species, which has a worldwide distribution, is placed in sect. *Laevispora*. *Asterostroma andinum* is primarily diagnosed by subglobose to globose basidiospores measuring  $6\text{--}7.5 \times 5\text{--}6.5\text{ }\mu\text{m}$  and asterosetal rays measuring  $30\text{--}130 \times 4\text{--}8\text{ }\mu\text{m}$ . The morphologically similar *Asterostroma laxum* Bres. produces smaller rays measuring up to  $40\text{ }\mu\text{m}$  in length (Parmasto 1970, Boidin et al. 1997).

The features distinguishing *Asterostroma* species reported from Japan can be found in the following key.

**Key to species of the genus *Asterostroma* in Japan**

- 1. Basidiospores smooth, subglobose ..... *A. andinum*
- 1. Basidiospores ornamented. .... 2
- 2. Basidiospores subglobose,  $4.8\text{--}6 \times 4\text{--}5\text{ }\mu\text{m}$  ..... *A. cervicolor*
- 2. Basidiospores subglobose to globose, larger (up to  $8 \times 8.5\text{ }\mu\text{m}$  or more), with subcylindrical and obtuse ornaments ..... 3
- 3. Basidiospores  $8.5\text{--}11 \times 7.5\text{--}9\text{ }\mu\text{m}$ ; basidiomata only on mangrove trees ..... *A. macrosporum*
- 3. Basidiospores smaller than  $8.5\text{--}11 \times 7.5\text{--}9\text{ }\mu\text{m}$ . .... 4
- 4. Basidia  $40\text{--}60 \times 6.5\text{--}8.5\text{ }\mu\text{m}$ ; basidiomata on monocotyledonous trees of angiosperms ..... *A. boninense*
- 4. Basidia  $18\text{--}41 \times 5\text{--}8.5\text{ }\mu\text{m}$ ; basidiomata both on angiosperms and gymnosperms ..... *A. muscicola*

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## MYCOTAXON

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Three new species of *Scytalidium* from soil

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**Abstract** — Three new species of dematiaceous hyphomycetes from soil in China, *Scytalidium nielamuense*, *S. verruculosum*, and *S. xigazense*, are described and illustrated. The type specimens (dried cultures) and living cultures are deposited in the Herbarium of Shandong Agricultural University Plant Pathology (HSAUP). Isotypes are kept in the Herbarium of the Institute of Microbiology, Academia Sinica (HMAS).

**Key words** — taxonomy, soil fungi

## Introduction

Since Pesante (1957) erected *Scytalidium* for *S. lignicola* Pesante, 22 species have been recognized worldwide (Index Fungorum 2010). This genus is characterized by dematiaceous, intercalary or terminal arthroconidia formed by fragmentation of undifferentiated hyphae. The arthroconidia are often thick-walled, smooth, occasionally verrucose in age, mid or dark brown, cylindrical, oblong, doliform or broadly ellipsoidal, often 0-septate, when septate with septa sometimes thick and very dark, often constricted at the septum; fission arthroconidia of a second type are hyaline, or pale to mid-brown, thin-walled, smooth, cylindrical, single-celled, truncate at each end. (Also refer to Ellis 1971.) During a recent survey of soil hyphomycetes in China, three new species of *Scytalidium* were found and are described below.

## Taxonomic descriptions

*Scytalidium nielamuense* Y.M. Wu & T.Y. Zhang, sp. nov.

FIG. 1

MYCOBANK MB 518543

*Coloniae in PDA effusae, plus minusve radiatim sulcatae, crassae. Mycelium aerium et immersum. Vegetativae hyphae laeves, subhyalinae vel brunneolae, ramosae, septatae, inflates cellulis 1.5–2 µm latae. Fertiles hyphae laeves, hyalinae vel subhyalinae, septatae,*

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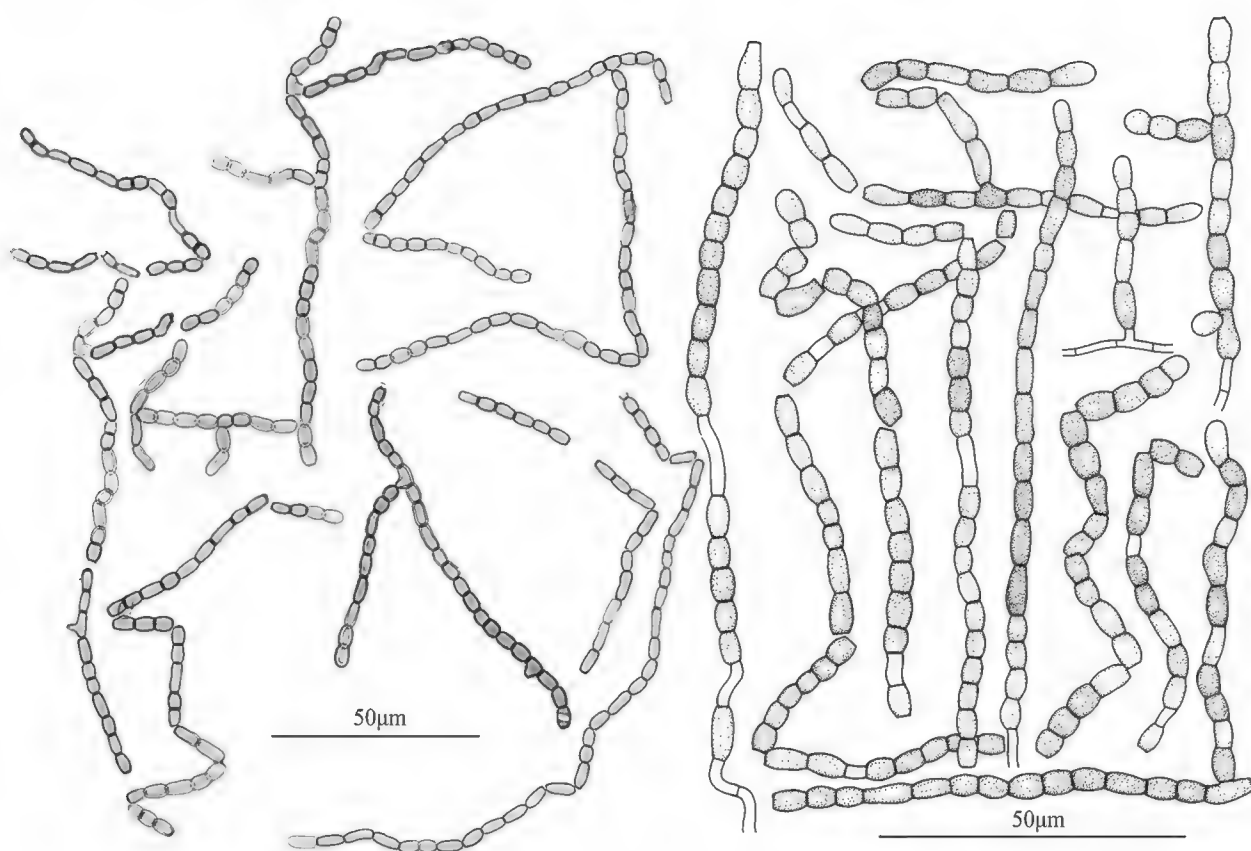


FIG. 1. Conidia and conidiogenous cells of *Scytalidium nielamuense* (ex holotype). Left: photomicrographs; right: drawings. (Bars = 50 µm).

*schizolitic secedentibus arthroconidis. Conidia subhyalina vel flavo-brunnea, cylindracea, oblongo-elliptica vel doliformia, laevia, 0-septata,  $3.8\text{--}7.5 \times 2.5\text{--}3.8$  µm.*

HOLOTYPE: China, Tibet, Nielamu, from a grassland soil, altitude 2250 m, 14 Sept. 2007, Y.M. Wu, HSAUPII<sub>07</sub>1268, **holotype**; HMAS 196252, **isotype**.

ETYMOLOGY: The epithet refers to the type location.

Colonies on PDA after two weeks at 25°C, effuse, growing slowly 2–3 cm diam., more or less radially folded, thick Mycelium partly superficial, partly immersed. Vegetative hyphae smooth, subhyaline to pale brown, branched, sparsely to regularly septate, sometimes slightly constricted at the septa, and often with individual cells rather variable in shape and slightly swollen, 1.5–2 µm wide; hyphae sometimes aggregating into strands. Fertile hyphae scarcely differentiated from vegetative hyphae, smooth, hyaline to subhyaline, with septa more closely spaced, fragmenting by schizolytic dehiscence to form arthroconidia. Conidia cylindrical to oblong-elliptical or doliform, vary in width depending on the parent hypha, subhyaline to yellow-brown, catenate, dry, simple, 0-septate, smooth,  $3.8\text{--}7.5 \times 2.5\text{--}3.8$  µm.

This fungus somewhat resembles *Scytalidium vaccinii* Dalpé et al. (Dalpé et al. 1989) in conidial morphology. However, the latter has larger ( $7\text{--}14 \times 3\text{--}4$  µm), guttulate conidia, which remain connected in zigzag chains.

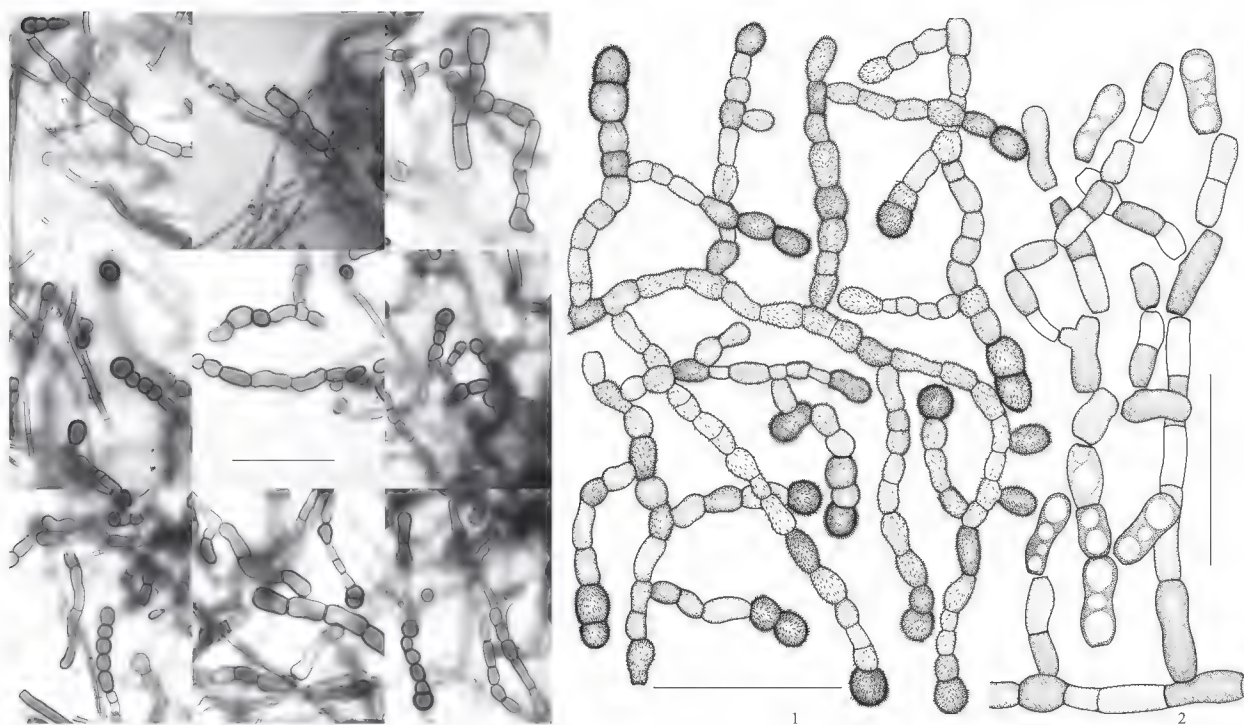


FIG. 2. Conidia and conidiogenous cells of *Scytalidium verruculosum* (ex holotype).  
Left: photomicrographs; right: drawings. (Bars = 50 µm).

***Scytalidium verruculosum* Y.M. Wu & T.Y. Zhang, sp. nov.**

FIG. 2

MYCOBANK MB 513017

*Coloniae in PDA effusae. Mycelium partim superficiale et partim in substrato. Hyphae ramosae, septatae, subhyalinae vel pallide brunneae, 2–4 µm latae. Conidia biformis: (1) cylindracea, catenata, sicca, simplicia, 0–1-septata, brunnea, incrassata, verrucosa, utroque truncata, interdum clavata vel pyriformia, basi truncata et apice rotundata, aegre secedentes, 8–20 × 5–10 µm; (2) clavata vel pyriformia, cateenata, sicca, simplicia, 0-septata, pallide-brunnea, tenuia et laevia, basi truncata et apice rotundata, facile fragmentantia, 10–26.5 × 6–9 µm.*

HOLOTYPE: China, Tibet, Zhangmu, from a mountain soil, altitude 2300 m, 14 Sept. 2007, Y.M. Wu, HSAUPII<sub>07</sub> 1328, **holotype**; HMAS 196253, **isotype**.

ETYMOLOGY: The epithet refers to the verrucose conidia of this species.

Colonies on PDA after two weeks at 25°C, effuse, growing very slowly, 2–3 cm diam., centre slightly raised, velvety, olivaceous brown. Mycelium partly superficial, partly immersed. Hyphae subhyaline to pale brown, smooth, septate, 2–4 µm thick, branched or unbranched. Conidia of two kinds: (1) cylindrical, catenate, dry, simple, 0–1-septate, medio-brown to dark brown, rough-walled, verrucose, truncate at both ends, sometimes clavate to pyriform, with a truncate base and rounded apex, not easily seceding, 8–20 × 5–10 µm; (2) clavate to pyriform, catenate, dry, simple, 0–catenate, dry, simple, 1-septate, pale-brown, thin and smooth-walled, with a truncate base and rounded apex, seceding schizolytically and easily, 10–26.5 × 6–9 µm.



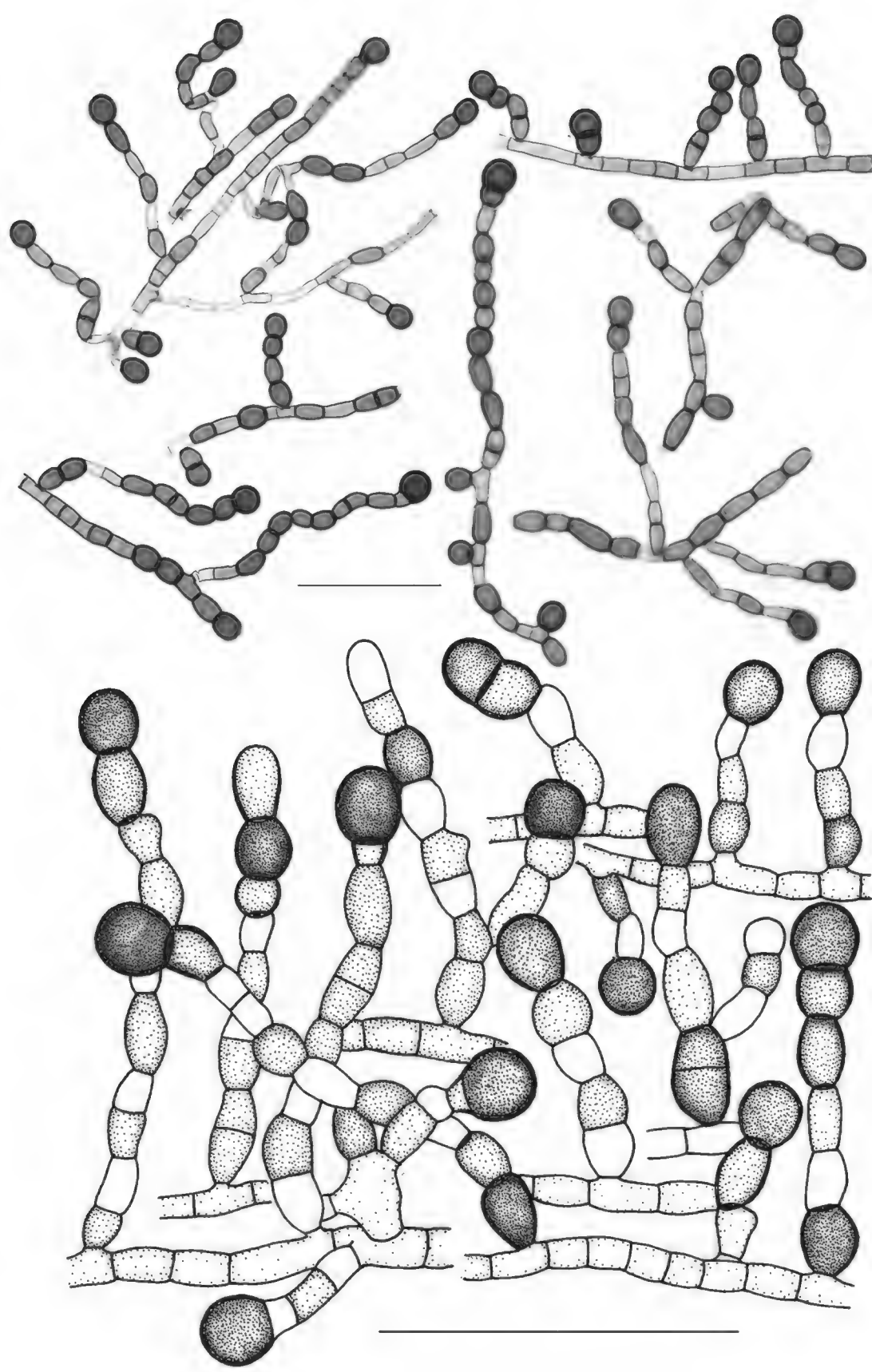


FIG. 3. Conidia and conidiogenous cells of *Scytalidium xigazense* (ex holotype) .  
Above: photomicrographs; below: drawings. (Bars = 50 μm).

This fungus somewhat resembles *Scytalidium infestans* Iwatsu et al. (Iwatsu et al. 1990) in conidial morphology. However, conidia of the latter are longer and narrower ( $4\text{--}30 \times 2\text{--}4.5 \mu\text{m}$ ), and rarely verrucose. *Scytalidium infestans* was described as a systemic pathogen of marine fish, whereas *S. verruculosum* is a soil fungus.

***Scytalidium xigazense* Y.M. Wu & T.Y. Zhang, sp. nov.**

FIG. 3

MYCOBANK MB 518397

*Coloniae in PDA effusae. Mycelium partim superficiale et partim in substrato. Hyphae ramosa, septata, subhyalina vel pallide brunnea, 1–3  $\mu\text{m}$  crassa. Conidia cylindracea, interdum clavata vel pyriformia, catenata, sicca, simplicia, 0–1-septata, subhyalina vel pallide brunnea, incrassata, laevia, utroque truncata, basi truncata et apice rotundata, 7–11  $\times$  4–8  $\mu\text{m}$ .*

HOLOTYPE: China, Tibet, Xigaze, from a mountain soil, altitude 3700 m, 7 Sept. 2007, Y.M. Wu, HSAUPII<sub>07</sub>0957, **holotype**; HMAS 196254, **isotype**.

ETYMOLOGY: The epithet refers to the type location.

Colonies on PDA after two weeks at 25°C, effuse, growing very slowly, 1.5–2.5 cm diam., centre slightly raised, velvety or floccose, olivaceous-gray. Mycelium partly superficial, partly immersed. Hyphae mostly subhyaline to pale brown, smooth, septate, 1–3  $\mu\text{m}$  thick, branched. Conidia cylindrical, sometimes clavate to pyriform, vary in width depending on the parent hypha, catenate, dry, simple, 0–1-septate, subhyaline to brown, smooth, truncate at both ends, or with a truncate base and rounded apex, not easily seceding, 7–11  $\times$  4–8  $\mu\text{m}$ .

This fungus somewhat resembles *Scytalidium fulvum* Morgan-Jones & Gintis (Morgan-Jones et al. 1984) in conidial morphology. However, conidia of the latter are longer and narrower (12–14  $\times$  2–3  $\mu\text{m}$ ).

### Acknowledgments

The authors are grateful for pre-submission comments and suggestions provided by Dr. Eric McKenzie, Prof. Y.L. Guo, and Dr. Shaun Pennycook. This project was supported by the National Science Foundation of China (no. 30670014 & 30499340).

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## MYCOTAXON

DOI: 10.5248/114.211

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**A new species of *Phellinus* (Hymenochaetaceae)  
growing on bamboo in tropical China**

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**Abstract** — *Phellinus bambusicola* sp. nov. is described and illustrated from Hainan Province, southern China. It has annual and resupinate basidiocarps, clay-buff to pale fawn pore surface, abundant hymenial setae, broadly ellipsoid and thin-walled basidiospores, setal hyphae present in the subiculum but absent at the sterile margin, and a growth on bamboo. The new species is similar to *Phellinus ferruginosus*, but the latter has an annual to perennial growth habit, yellowish brown to dark reddish brown pore surface, smaller pores (6–8 per mm), setal hyphae present at the sterile margin, and narrowly ellipsoid basidiospores.

**Key words** — *Hymenochaetales*, polypore, taxonomy

**Introduction**

*Phellinus* Quél., with over 250 taxa worldwide, is the largest genus in the *Hymenochaetaceae* (Larsen & Cobb-Poulsen 1990, Dai 1999, 2010, Núñez & Ryvarden 2000, Gibertoni et al. 2004, Ryvarden 2004, Parmasto 2007). Wanger & Fischer (2002), who studied *Phellinus* sensu lato and *Inonotus* sensu lato phylogenetically, divided the *Phellinus*–*Inonotus* complex into 13 genera. Since Dai (1999) recorded 45 species of *Phellinus* from East Asia new species or new records have been found in China, where about 50 species in the genus have

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been reported thus far (Dai 1995, 1999, Dai et al. 2003, 2008, Dai & Yang 2008, Cui et al. 2009).

During a study of wood-inhabiting fungi in southern China, an unknown species of *Phellinus* growing on bamboo was identified and is described in the present paper.

### Materials and methods

The studied specimens were deposited in herbaria as cited below. The microscopic procedure follows Cui & Dai (2008). In presenting the variation in the size of the spores, 5% of measurements were excluded from each end of the range, and given in parentheses. In the text the following abbreviations are used: IKI = Melzer's reagent, IKI- = negative in Melzer's reagent, KOH = 5% potassium hydroxide, CB = Cotton Blue, CB- = acyanophilous, L = mean spore length (arithmetic average of all spores), W = mean spore width (arithmetic average of all spores), Q = variation in the L/W ratios between the specimens studied, n = number of spores measured from given number of specimens. Sections were studied at magnification up to  $\times 1000$  using a Nikon Eclipse E 80i microscope and phase contrast illumination. Drawings were made with the aid of a drawing tube. Special colour terms follow Anonymous (1969) and Petersen (1996).

### Taxonomy

*Phellinus bambusicola* L.W. Zhou & B.S. Jia, sp. nov.

FIG. 1

MYCOBANK MB 518776

*Carpophorum annuum*, *resupinatum*. *Facies pororum avellanea vel hinnulea*; *pori angulati*, 3–5 per mm. *Systema hypharum dimiticum*, *hyphae generatoriae septatae, efibulatae*. *Sporae late ellipsoideae*, IKI-, CB-,  $4.2\text{--}5 \times 3.1\text{--}4\ \mu\text{m}$ .

TYPE. — China. Hainan Province, Changjiang County, Bawangling Nature Reserve, on dead bamboo, 8.XII.2009 Cui 8692 (holotype in BJFC, isotype in IFP).

ETYMOLOGY — *bambusicola* (Lat.): refers to growth on bamboo.

FRUITBODY — Basidiocarps annual, resupinate, firmly attached to the substrate, not readily separable, without odour or taste when fresh, hard corky when dry, up to 15 cm long, 5 cm wide and 2 mm thick at centre; sterile margin pale clay-buff to pale fawn, up to 3 mm wide. Pore surface clay-buff to pale fawn when dry; pores angular, 3–5 per mm, dissepiments thin, entire when juvenile, lacerate with age. Subiculum yellowish brown to fawn-brown, hard corky, about 0.4 mm thick. Tubes concolorous with pore surface, corky, about 1.6 mm long.

HYPHAL STRUCTURE — Hyphal system dimitic; all septa without clamp connections; skeletal hyphae IKI-, CB-; tissue darkening but otherwise unchanged in KOH.



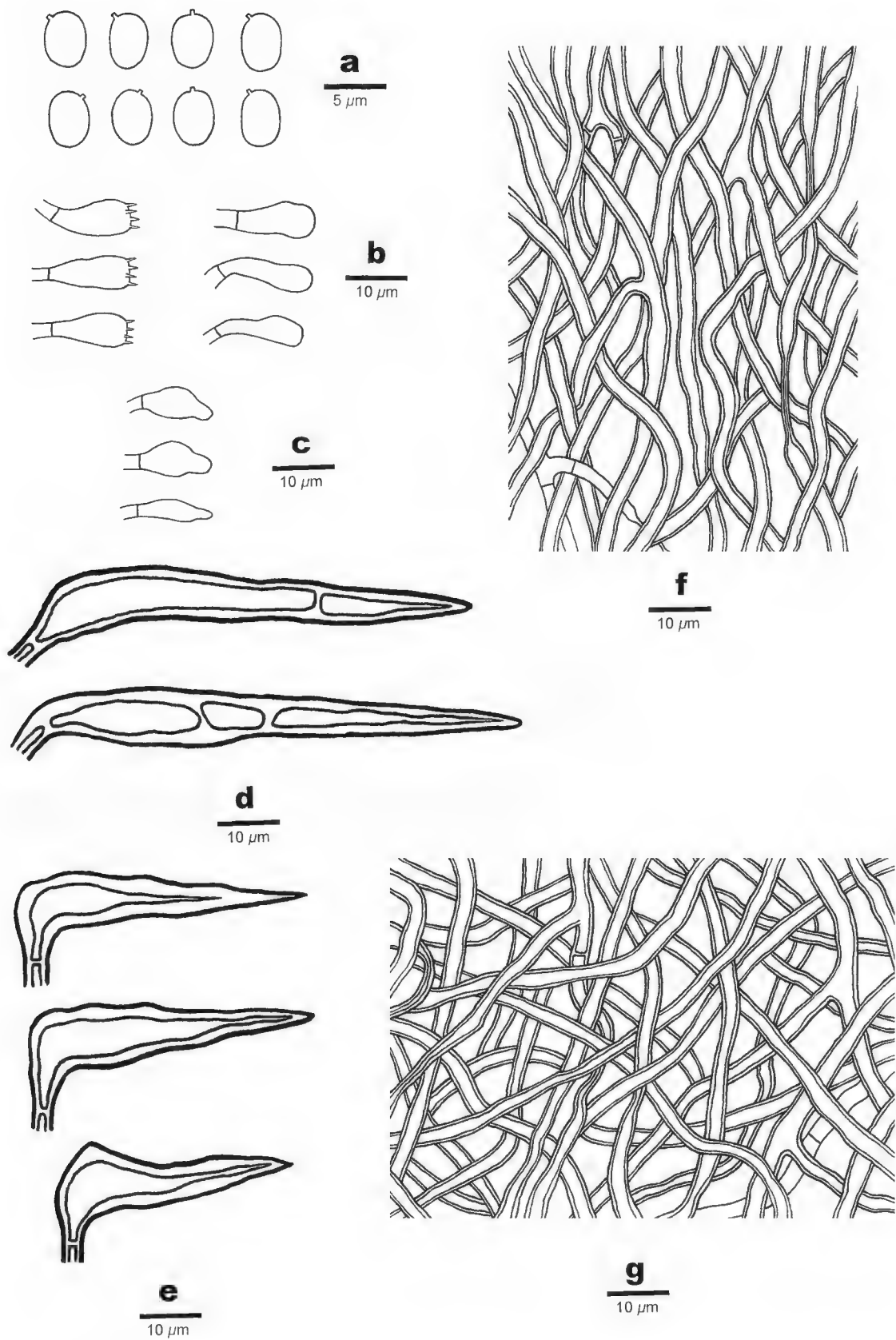


FIG. 1. Microscopic structures of *Phellinus bambusicola* (drawn from the holotype).  
a: Basidiospores. b: Basidia and basidioles. c: Cystidioles.  
d: Hyphoid setae. e: Setae. f: Hyphae from trama. g: Hyphae from context.

**SUBICULUM** — Generative hyphae infrequent, hyaline to pale yellowish, thin- to slightly thick-walled, occasionally branched, some collapsed, 2.2–3.5  $\mu\text{m}$  in diam; skeletal hyphae pale yellowish to apricot-orange, thick-walled with a wide lumen, occasionally branched, some collapsed, interwoven, 2–5  $\mu\text{m}$  in diam; setal hyphae frequent, apricot-orange, thick-walled, tapering to apex, 6.5–10.5  $\mu\text{m}$  wide and up to 110  $\mu\text{m}$  long.

**TUBES** — Generative hyphae infrequent, hyaline to pale yellowish, thin- to thick-walled, frequently branched and some collapsed, 1.8–3.5  $\mu\text{m}$  in diam; skeletal hyphae dominant, pale yellowish to apricot-orange, thick-walled, occasionally branched, and some collapsed, parallel along the tubes, 1.6–4  $\mu\text{m}$  in diam. Hymenial setae frequent, ventricose to subulate, tapering to apex, dark brown, thick-walled, 31.8–54.5  $\times$  9.2–14.3  $\mu\text{m}$ . Cystidia absent, fusoid cystidioles present, hyaline, thin-walled, 9.8–17  $\times$  4.9–6.3  $\mu\text{m}$ . Basidia clavate, bearing four sterigmata and a simple septum at the base, 8.7–18  $\times$  3.9–6  $\mu\text{m}$ ; basidioles in shape similar to basidia, but slightly smaller. Irregular crystals present in trama and hymenia.

**SPORES** — Basidiospores broadly ellipsoid, hyaline, thin-walled, smooth, IKI–, CB–, (4–)4.2–5(–5.9)  $\times$  (3–)3.1–4  $\mu\text{m}$ ,  $L = 4.68 \mu\text{m}$ ,  $W = 3.56 \mu\text{m}$ ,  $Q = 1.31$  ( $n = 30/1$ ).

**TYPE OF ROT** — White rot.

**REMARKS** — *Phellinus bambusicola* was found on bamboo in tropical China. It is characterized by annual, resupinate basidiocarps, a clay-buff to pale fawn pore surface, abundant hymenial setae, broadly ellipsoid and thin-walled basidiospores, setal hyphae present in the subiculum while absent at the sterile margin, and growth on bamboo.

This species is similar to *Phellinus ferruginosus* (Schrad.) Pat., but the latter shows an annual to perennial growth habit, yellowish brown to dark reddish brown pore surface, smaller pores (6–8 per mm, Dai 1999), setal hyphae present at the sterile margin, and narrowly ellipsoid basidiospores are (4.7–5.3  $\times$  3.0–3.5  $\mu\text{m}$ ,  $L = 5.04 \mu\text{m}$ ,  $W = 3.16 \mu\text{m}$ ,  $Q = 1.59$ ).

*Phellinus bambusarum* (Rick) M.J. Larsen also grows on bamboo and may be confused with *P. bambusicola*. However, *P. bambusarum* differs by a perennial growth habit, smaller pores (6–8 per mm) with thick-walled dissepiments, rare and smaller hymenial setae (13–25  $\times$  6–8  $\mu\text{m}$ ), and globose to subglobose and dextrinoid basidiospores (Ryvarden 2004).

*Phellinus bambusinus* (Pat.) Pat., another species growing on bamboo, is distinguished from *P. bambusicola* in its pileate basidiocarps, ochraceous brown and glancing (reflective) pore surface, small invisible pores, and ovoid basidiospores (5  $\times$  4  $\mu\text{m}$ ); moreover, it has conidia (Larsen & Cobb-Poullé 1990).

## A key to species of *Phellinus* on bamboo

1. Basidiocarps pileate; conidia present ..... *P. bambusinus*
1. Basidiocarps resupinate; conidia absent ..... 2
2. Pores 6–8 per mm; basidiospores subglobose, dextrinoid ..... *P. bambusarum*
2. Pores 3–5 per mm; basidiospores broadly ellipsoid, IKI– ..... *P. bambusicola*

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We express our gratitude to Drs. Wjacheslav Spirin (St. Petersburg, Russia) and Zheng Wang (Yale University, USA) who reviewed the manuscript. The research was financed by the National Natural Science Foundation of China (Project No. 30910103907) and the Ministry Science and Technology of China (Project No. 2006FY110500-5).

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## MYCOTAXON

DOI: 10.5248/114.217

Volume 114, pp. 217–223

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**Two new species of *Septobasidium* (Septobasidiaceae)  
from Hainan Province in China**CHUNXIA LU<sup>1,2</sup> & LIN GUO<sup>1\*</sup>

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**Abstract** — Two new species, *Septobasidium hainanense* on *Harpullia* sp. associated with *Pseudaulacaspis* sp. and *Septobasidium ligustri* on *Ligustrum sinense* associated with *Lepidosaphes* sp., are described.

**Key words** — *Pucciniomycetes*, *Septobasidiales*, taxonomy

The mycota is very rich in tropical forests of Hainan. Several mycological investigations dealing with many new species including the genus *Septobasidium* were published recently (Dai & Cui 2006, Dai & Li 2010, Cui et al. 2009, Dai et al. 2009, Lu & Guo 2009a, 2010b, Yuan & Dai 2008, Xiong & Dai 2008, Wei & Dai 2008). The present paper belongs to a series of studies devoted to the fungal diversity of the Hainan Province. Two new species of *Septobasidium* are described as follows:

***Septobasidium hainanense* C.X. Lu & L. Guo, sp. nov.**

FIGS. 1–7

MYCOBANK MB 518658

*Basidiomata resupinata, 0.2–2.5 cm longa, 0.15–1 cm lata, purpurea, margine determinata, superficie laevia, in sectione 220–830 µm crassa. Subiculum brunneum, 25–60 µm crassum. Columnae brunneae, 50–110 µm altae, 60–155 µm crassae vel hyphis laxae completae. Strata hyphararum 70–505 µm alta, saepe strata horizontalia formantia, interdum hyphae partim successiveque crescentes et texturam hemisphaericam tum formantes. Hymenium 50–200 µm crassum. Hyphae hymenii erectae. Basidia cylindrica, recta vel curvata, 4-cellularia, 25–36 × 7–13 µm, hyalina vel brunneola. Sine probasidio. Basidiosporae non visae. Haustoria ex hyphis irregulariter spiralibus constantia.*

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\*corresponding author



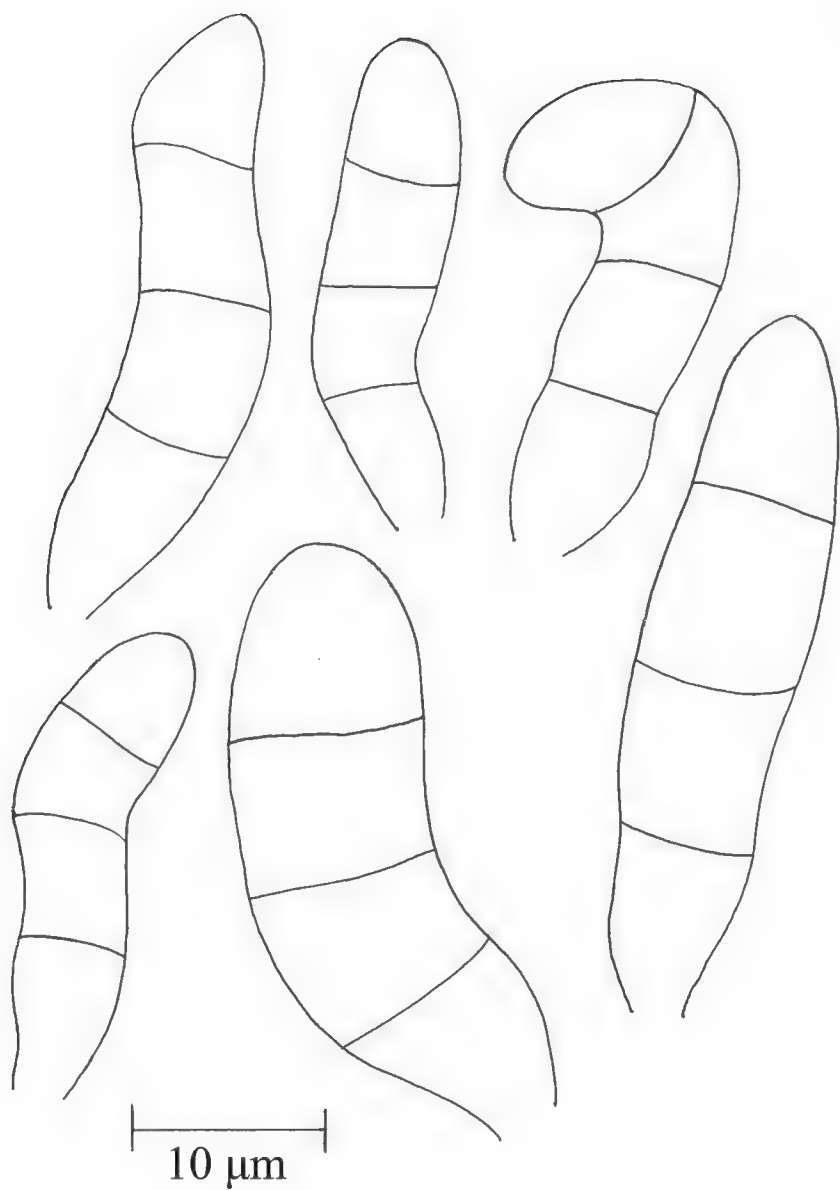
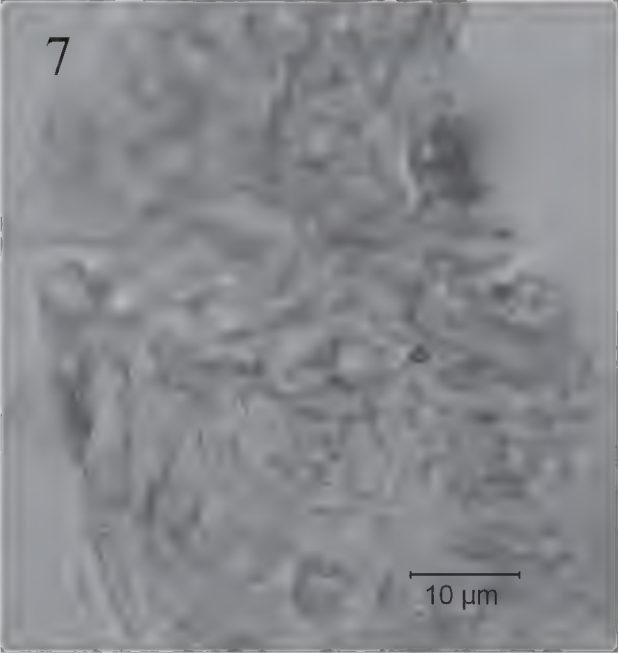
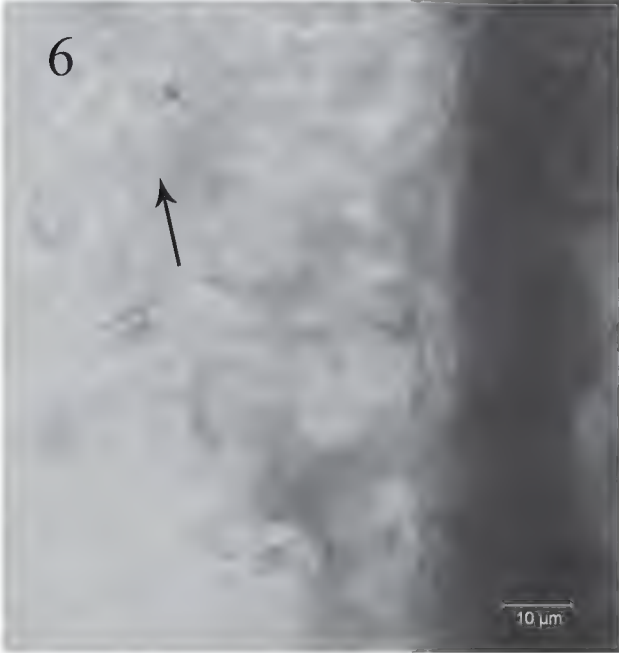
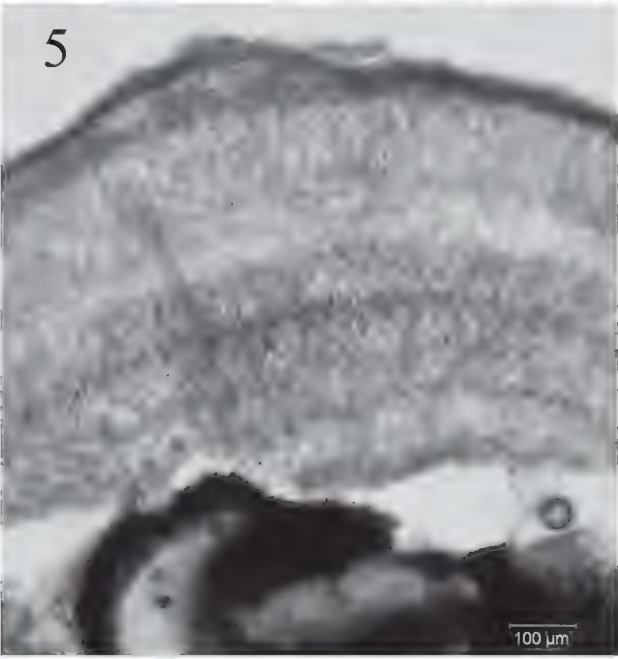
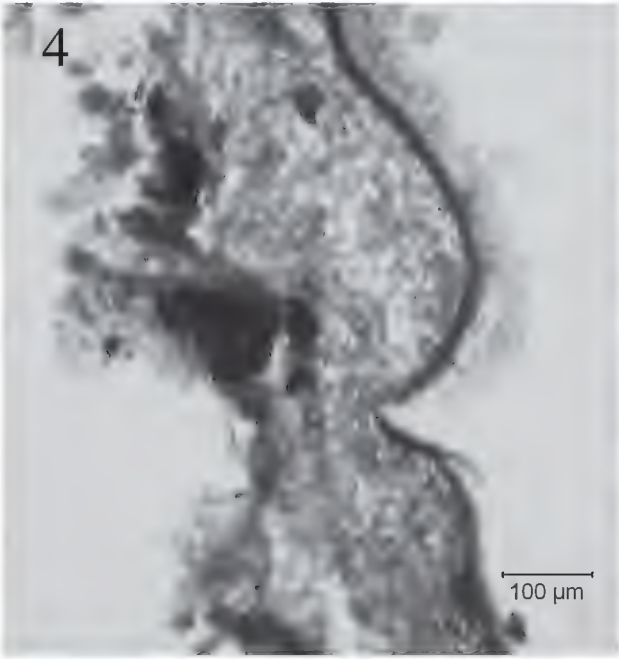
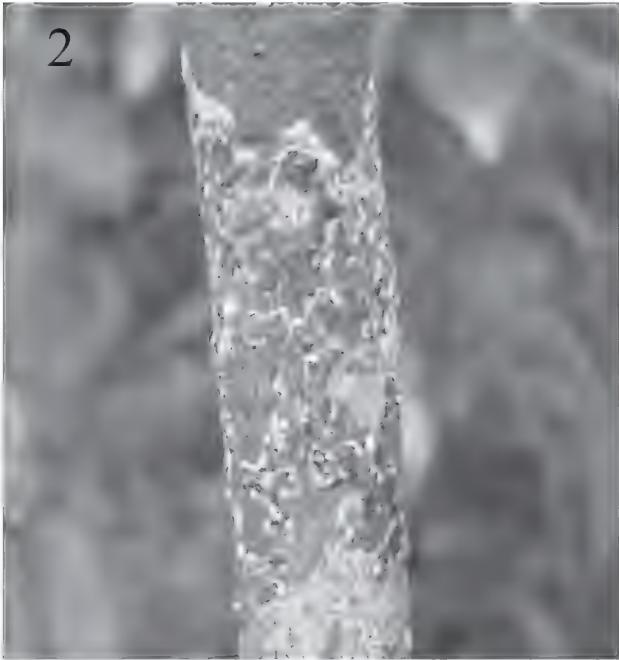


FIG. 1. Basidia of *Septobasidium hainanense* (HMAS 240078, holotype).

TYPE: On *Harpullia* sp. (*Sapindaceae*): China, Hainan, Bawangling, Yajia, alt. 740 m, 12.XII.2009, Y.F. Zhu & L. Guo 141, HMAS 240078 (**holotype**), associated with *Pseudaulacaspis* sp. (*Diaspididae*).

Basidiomata on trunks, resupinate, small, rounded, elongate or irregular, often confluent, 0.2–2.5 cm long, 0.15–1 cm wide, purple; margin determinate; surface smooth, often with mounds. In section 220–830 μm thick. Subiculum brown, 25–60 μm thick. Pillars brown, 50–110 μm high, 60–155 μm wide, sometimes loosely filled with hyphae from the subiculum. Hyphal layer 70–505 μm high, often forming a distinct horizontal layer, sometimes hyphae partly and successively growing and forming hemispheric tissue. Hymenial layer 50–200 μm thick, with closely arranged upright hyphae. Basidia arising directly from

FIGS. 2–7 (right). *Septobasidium hainanense* (HMAS 240078, holotype). 2. Basidiomata on trunk. 3–5. Sections of basidiomata. 6. Basidium (arrow). 7. Haustoria.



the hyphae, cylindrical, straight or curved, 4-celled,  $25\text{--}36 \times 7\text{--}13\ \mu\text{m}$ , hyaline or brownish, without a probasidial cell. Basidiospores not seen. Haustoria consisting of irregularly coiled hyphae.

REMARKS: Morphologically, *Septobasidium hainanense* is similar to *S. lichenicola* (Berk. & Broome) Petch, from which it differs in having small patches of basidiomata, hyphae partly growing and forming hemispheric tissue, and with pillars or loosely filled with hyphae from subiculum. *Septobasidium lichenicola* has large patches of basidiomata, hyphae not forming hemispheric tissue and not loosely filled with hyphae from subiculum.

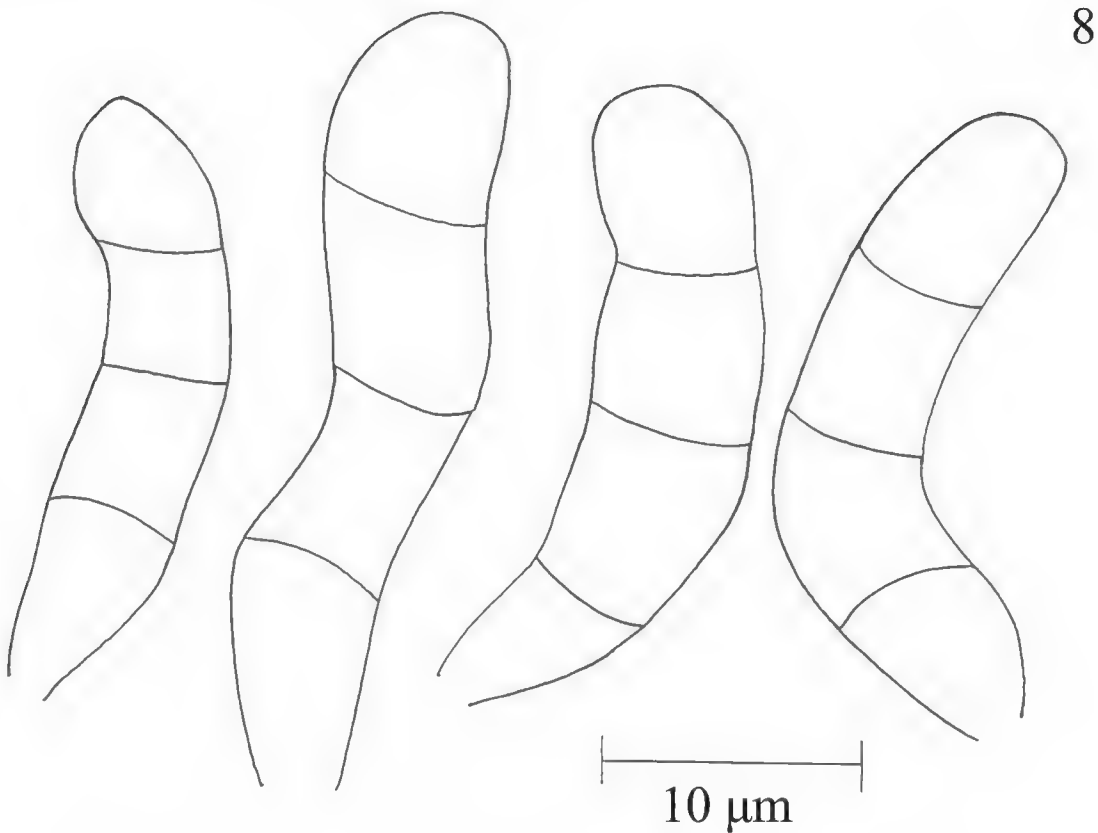


FIG. 8. Basidia of *Septobasidium ligustri* (HMAS 240079, holotype).

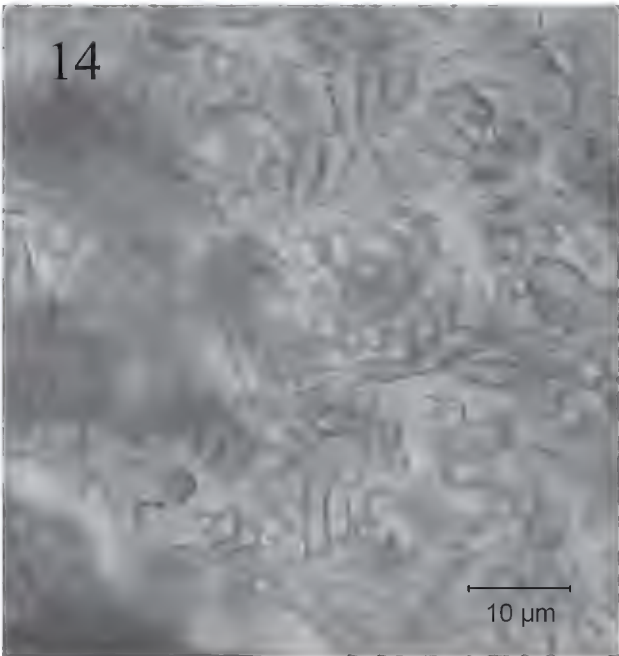
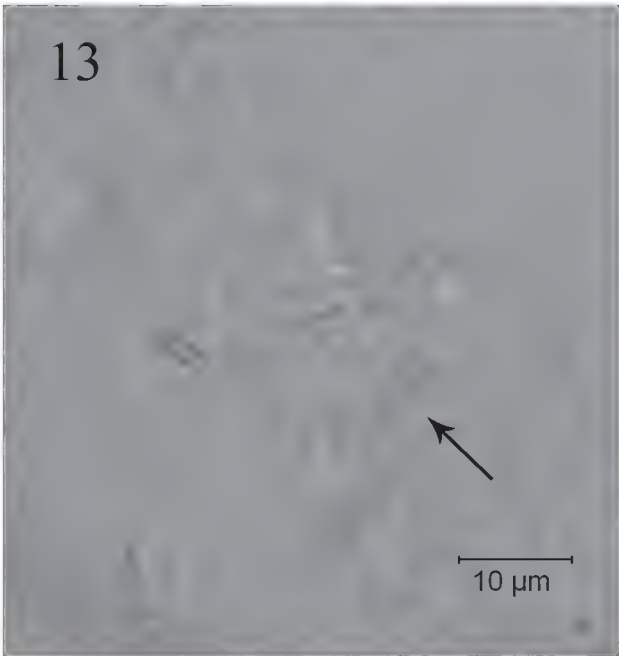
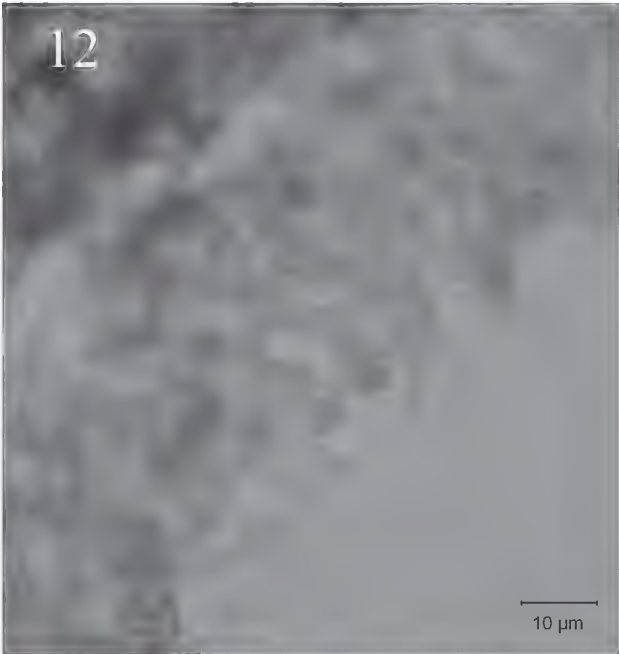
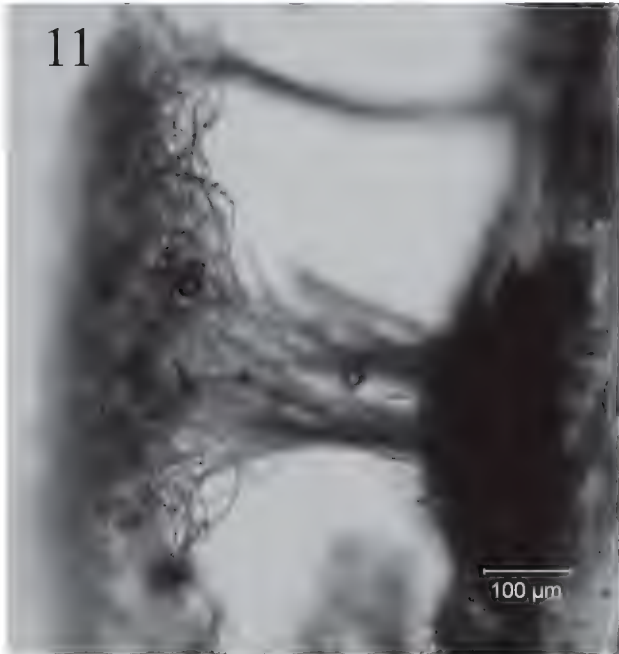
*Septobasidium ligustri* C.X. Lu & L. Guo, sp. nov.

FIGS. 8–14

MYCOBANK MB 518659

*Basidiomata resupinata, 9–20 cm longa, 1–3 cm lata, griseo-brunnea, margine determinata, superficie laevia, maturitate fissurata, in sectione 480–630 μm crassa. Subiculum brunneum, 20–60 μm crassum. Columnae brunneolae, 210–390 μm altae, 30–150 μm latae, extus ramosae strata hyphararum 100–170 μm alta tum formantes. Hymenium 50–80 μm crassum. Hyphae hymenii irregulariter dispositae, erectae, ramosae. Basidia cylindrica, recta vel curvata, 4-cellularia,  $15\text{--}29 \times 5\text{--}7.5\ \mu\text{m}$ , hyalina. Sine probasidio. Sterigmata 3–8 μm longa. Basidiospora ovoidea,  $9 \times 4\ \mu\text{m}$ , hyalina. Haustoria ex hyphis irregulariter spiralibus constantia.*

FIGS. 9–14 (right). *Septobasidium ligustri* (HMAS 240079, holotype). 9. Basidiomata on branch. 10–11. Sections of basidiomata. 12. Hymenium. 13. Basidium (arrow). 14. Haustoria.





TYPE: On *Ligustrum sinense* Lour. (*Oleaceae*): China, Hainan, Wanning, Xinglong Tropical Plant Garden, alt. 38 m, 6.XII.2009, Y.F. Zhu & L. Guo 41, HMAS 240079 (holotype), associated with *Lepidosaphes* sp. (*Diaspididae*).

Basidiomata on branches, resupinate, 9–20 cm long, 1–3 cm wide, grey-brown; margin determinate; surface smooth, becoming cracked. In section 480–630  $\mu\text{m}$  thick. Subiculum brown, 20–60  $\mu\text{m}$  thick. Pillars brownish, 210–390  $\mu\text{m}$  high, 30–150  $\mu\text{m}$  wide, branched outwards to form a 100–170  $\mu\text{m}$  high hyphal layer. Hymenium 50–80  $\mu\text{m}$  thick, with irregularly arranged upright branched hyphae. Basidia arising directly from the hyphae, cylindrical, straight or curved, 4-celled,  $15\text{--}29 \times 5\text{--}7.5 \mu\text{m}$ , hyaline, without a probasidial cell. Sterigmata 3–8  $\mu\text{m}$  long. Basidiospore ovoid,  $9 \times 4 \mu\text{m}$ , hyaline. Haustoria consisting of irregularly coiled hyphae.

REMARKS: Morphologically, *Septobasidium ligustri* is similar to *S. septobasidioides* (Henn.) Höhn. & Litsch., but differs mainly in having grey-brown basidioma, thinner section (480–630  $\mu\text{m}$  vs about 1 mm) and smaller basidia ( $15\text{--}29 \times 5\text{--}7.5 \mu\text{m}$  vs  $40\text{--}55 \times 8.4\text{--}10 \mu\text{m}$ ).

To date, 28 species of *Septobasidium* have been reported in China (Sawada 1933, Couch 1938, Teng 1963, Tai 1979, Kirschner & Chen 2007, Lu & Guo 2009a, b, c, 2010a, b, Lu et al. 2010), including the two new species reported in this paper.

### Acknowledgements

The authors would like to express their deep thanks to Drs Eric H.C. McKenzie (Auckland, New Zealand) and Shuanghui He (Beijing Forestry University) for serving as pre-submission reviewers, to Dr. Shaun Pennycook (Auckland, New Zealand) for nomenclatural review, to Prof. Jianyun Zhuang (Institute of Microbiology, Chinese Academy of Sciences) for Latin corrections, to Mr Ziyu Cao (Institute of Botany, Chinese Academy of Sciences) for identifying the host plants, to Prof. Sanan Wu (Beijing Forestry University) for identifying the scale insects, and to Mrs Xiangfei Zhu for inking in line drawings. This study was supported by the Ministry of Science and Technology of the People's Republic of China (No. 2006FY110500–5).

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# MYCOTAXON

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## New records of smut fungi. 3

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**Abstract** — Three rare species of smut fungi are reported for the first time from the following areas: *Anthracoidea ortegae* from the Falkland Islands, *Entorrhiza casparyana* var. *casparyana* from Egypt, on a new host, *Juncus hybridus*, and *Haradaea moenchiae-manticae* from UK.

**Key words** — *Anthracoideaceae*, *Entorrhizaceae*, *Microbotryaceae*, taxonomy, *Ustilaginomycetes*

## Introduction

In this article, records of three rare species of smut fungi, *Anthracoidea ortegae*, *Entorrhiza casparyana* var. *casparyana*, and *Haradaea moenchiae-manticae*, are reported from new localities. The collections on which these records are based were obtained during visits to the herbaria at the Royal Botanic Garden Edinburgh (E) and the Royal Botanic Gardens, Kew (K, K(M)) in May 2010.

## Material and methods

Material from the herbaria of the Royal Botanic Garden Edinburgh (E) and the Royal Botanic Gardens, Kew [K and K(M)] was examined by light microscope (LM) and scanning electron microscope (SEM). For LM observations, the

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spores were mounted in lactophenol solution on glass slides, gently heated to boiling point and then cooled. The measurements of spores are given in the form: min–max (mean  $\pm$  1 standard deviation). For SEM, the spores were attached to specimen holders by double-sided adhesive tape and sputter coated with gold. The surface structure of spores was observed and photographed at 10 kV using a JEOL SM-6390 scanning electron microscope. The descriptions given below are based entirely on the specimens examined.

### New records

*Anthracoidea ortegae* Kukkonen, in Roivainen, Karstenia 17: 4, 1977. FIGS 1–2

SPECIMENS EXAMINED — On *Carex caduca* var. *ortegae* (Phil.) Kük.: Falkland Islands, West Falkland, Channel Hills, 1909–1911, leg. E. Vallentin (K 367 916); East Falkland, Darwin Harbour, 16 February 1908, leg. C. Skottsberg (K 367 906); East Falkland, Eliza Cove, Stanley Common, January 1938, leg. B.F., no. 49 (K(M) sine num.).

SORI in ovaries, scattered in the inflorescence, as broadly ellipsoidal or ovoid, black, hard bodies, 1.5–2 mm long, when young covered by a thin, whitish membrane; later becoming exposed but partly hidden by the glumes; mature sori powdery on the surface. SPORES irregularly polyangular, sometimes with protuberances, in plane view 14–19.5  $\times$  12.5–17.5 (16.9 $\pm$ 1.1  $\times$  15.2 $\pm$ 1.0)  $\mu$ m (n = 100), in side view 10–12.5  $\mu$ m thick, reddish brown; wall unevenly thickened, 1–2 (–2.5)  $\mu$ m thick, thickest at the angles, some spores with 1–3 indistinct internal swellings, some spores with light-refractive areas, verruculose.

DISTRIBUTION — On *Cyperaceae*: *Carex* (subgen. *Primocarex*, sect. *Unciniiformes*), South America (Argentina), South Atlantic Islands (Falkland Islands).

COMMENT — *Anthracoidea ortegae* was previously known only from the type locality: Argentina, Tierra del Fuego, Baliza, Ushuaia, 54°48' S, 68°12' W, on the same host plant (Roivainen 1977).

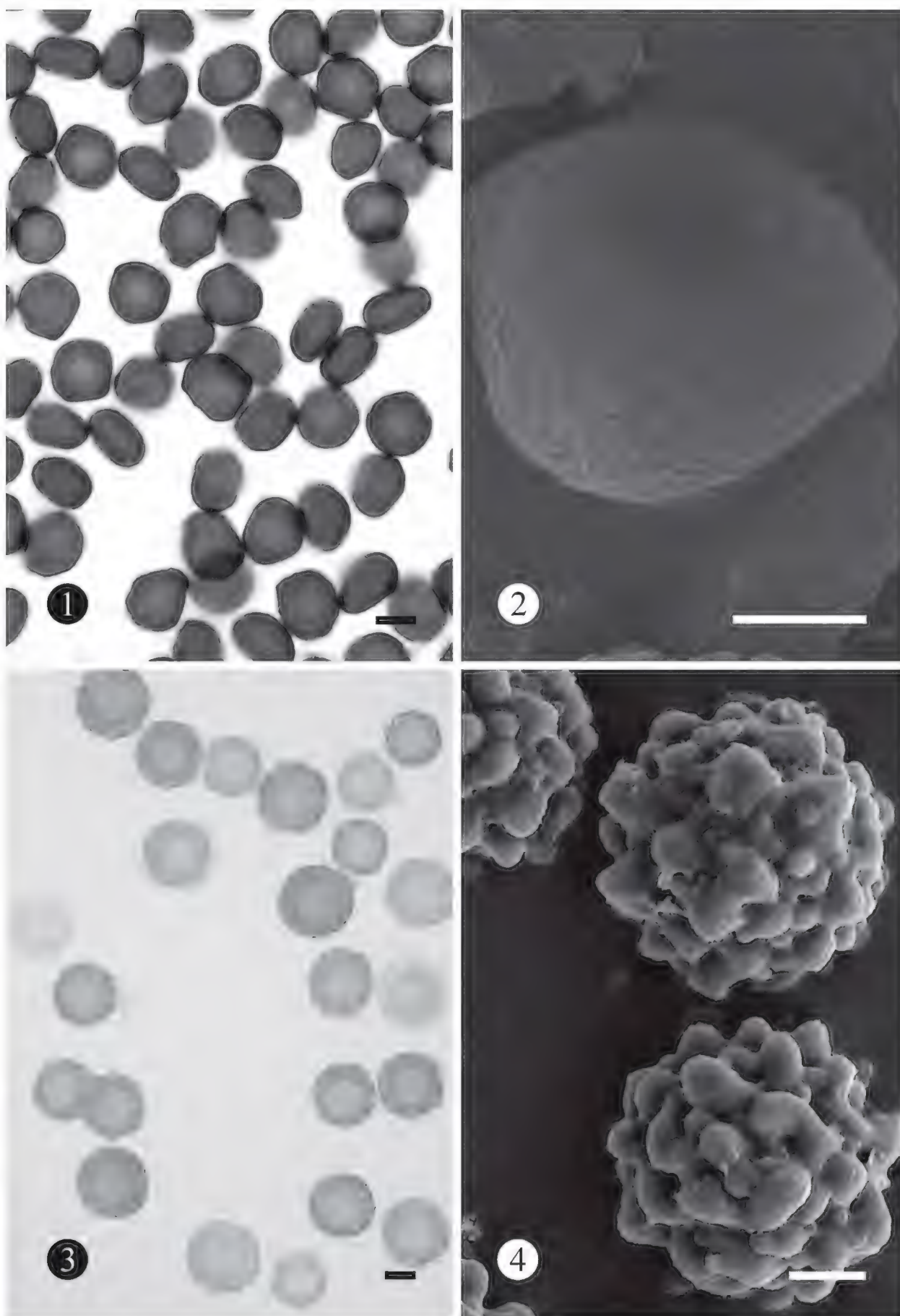
*Entorrhiza casparyana* (Magnus) Lagerh. var. *casparyana*, Hedwigia 27(9–10):

262, 1888.

FIGS 3–4

SPECIMEN EXAMINED — On *Juncus hybridus* Brot. (det. G. Snogerup): Egypt, “prope Nafeh in Arabia”, 16 May 1835, leg. W. Schimper (as *Juncus foliosus* Desf.), Unio itiner. 1835, no. 113 (E 352 462).

SORI on the roots forming elongated galls, filled with intracellularly developing spores. GALLS 4–6 mm long, brown. SPORE MASS granular. SPORES usually solitary, sometimes in pairs, globose or subglobose, 16.5–28  $\times$  15–26 (21.9 $\pm$ 2.0  $\times$  20.6 $\pm$ 1.9)  $\mu$ m (including ornamentation) (n = 100), occasionally some spores reach up to 32  $\mu$ m in length, subhyaline, light yellow or yellowish brown; in LM, wall two-layered, the inner layer 0.5–1.5  $\mu$ m thick, the outer layer variable in thickness (0.5–8  $\mu$ m, including ornamentation); variable in ornamentation, tuberculate or verrucose.



FIGS 1–2. Spores of *Anthracoidea ortegae* on *Carex caduca* var. *ortegae* in LM and SEM.  
 FIGS 3–4. Spores of *Entorrhiza casparyana* var. *casparyana* on *Juncus hybridus* in LM and SEM.  
 Scale bars: 1, 3 = 10  $\mu\text{m}$ , 2, 4 = 5  $\mu\text{m}$ .



DISTRIBUTION (of var. *caspariana*) — On *Juncaceae*: *Juncus alpino-articulatus* Chaix, *J. alpinus* Vill., *J. arcticus* Willd., *J. articulatus* L. (*J. lampocarpus* Ehrh. ex Hoffm.), *J. bufonius* L., *J. bulbosus* L., *J. caespiticius* E. Mey., *J. compressus* Jacq., ? *J. conglomeratus* L., *J. effusus* L., *J. geniculatus* Schrank, *J. gregiflorus* L.A.S. Johnson, *J. hybridus*, *J. inflexus* L., *J. planifolius* R. Br., *J. tenageia* Ehrh. ex L. f., *J. thomasi* Ten., Africa (Egypt, South Africa), Australasia (Australia, New Zealand), Europe (Bulgaria, Czech Republic, Denmark, including Faeroe Islands, Finland, France, Germany, Italy including Sardinia, Norway, Poland, Romania, Russia, Sweden, Switzerland, UK), North America (Canada) (Fineran 1978, Vánky 1994, Denchev & Minter 2008, Vánky & Shivas 2008). Records on four other hosts (*Eriophorum vaginatum* L. (*Cyperaceae*), *Juncus atricapillus* Drejer, *J. filiformis* L., and *J. squarrosus* L.) were treated by Fineran (1978) as doubtful or as later misinterpretations.

COMMENTS — In Africa, *Entorrhiza caspariana* has been previously known only from South Africa. Explanations about the possible situation of the locality 'Nafeh', where this plant specimen (Unio itiner. 1835, no. 113) was collected, can be found in Kirschner et al. (2004: 374): "The locality 'Nafeh' was not safely identified. W. Schimper, from late March, 1835, collected plants in the region around the monastery of St. Catharina at the foot of Mt Sinai [Dayr al Qiddisah Katrina]. ... Nafeh is therefore expected to be in that region, too."

*Entorrhiza caspariana* var. *tenuis* Denchev & H.D. Shin differs from typical *E. caspariana* in the following two respects: shorter spores (11.5–20 (–21.5)  $\mu\text{m}$  long) and shorter sori (1.2–3 mm long while the typical variety possesses sori up to 15 mm long) (Denchev et al. 2007). It is distributed on *Juncus tenuis* Willd. and currently known from Korea, Austria, Romania, and Costa Rica. Four species of *Entorrhiza* are known on *Juncus*: *E. aschersoniana* (Magnus) Lagerh. (Europe, Central America, and New Zealand), *E. caricicola* Ferd. & Winge (Europe and New Zealand), *E. caspariana*, and *E. casparianella* Vánky (New Zealand). A key to known *Entorrhiza* taxa on *Juncus* is given in Denchev & Minter (2008).

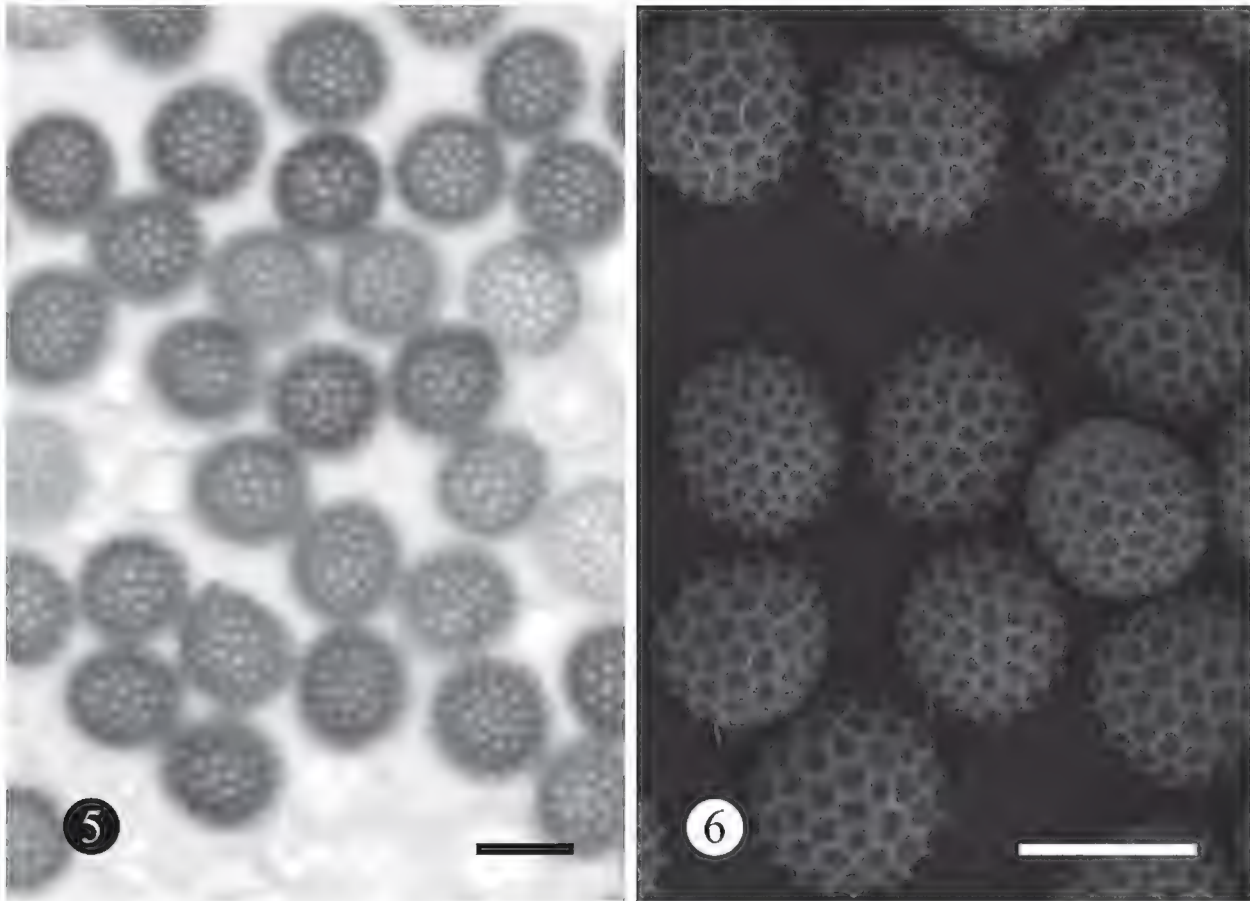
***Haradaea moenchiae-manticae* (Lindtner) Denchev & H.D. Shin, in Denchev et al., Mycologia Balcanica 3: 72, 2006. FIGS 5–6**

= *Ustilago moenchiae-manticae* Lindtner, Bulletin du Muséum d'Histoire Naturelle du Pays Serbe, Série B 3–4: 32, 1950.

= *Microbotryum moenchiae-manticae* (Lindtner) Vánky, Mycotaxon 67: 46, 1998.

SPECIMEN EXAMINED — On *Moenchia erecta* (L.) P. Gaertn. et al.: UK, Wales, Montgomeryshire, Ffridd Faldwyn, 15 May 1998, leg. A. Jones (as *Ustilago ? duriaeana*) (K(M) 106 303).

SORI destroying the ovules and filling the capsules with powdery, purplish chestnut spore mass. SPORES globose or subglobose, rarely broadly ellipsoidal,  $11\text{--}15.5 \times 10\text{--}13.5$  ( $13.0 \pm 0.8 \times 12.1 \pm 0.7$ )  $\mu\text{m}$  ( $n = 50$ ), purplish brown; reticulate, 6–7 meshes per spore diameter, meshes irregularly polyangular (pentagonal or hexagonal), 1.2–2.7  $\mu\text{m}$  long, muri (0.7–) 1.0–1.4  $\mu\text{m}$  high; in SEM the meshes often with a hemispherical protuberance on the bottom.



FIGS 5–6. Spores of *Haradaea moenchiae-manticae* on *Moenchia erecta* in LM and SEM. Scale bars = 10  $\mu$ m.

**DISTRIBUTION** — On *Caryophyllaceae*: *Moenchia erecta* (Bulgaria and UK), *M. mantica* (L.) Bartl. subsp. *mantica* (Romania and Serbia), Europe (Lindtner 1950, Vánky 1985, Denchev 1997).

**COMMENT** — *Haradaea moenchiae-manticae* is a new species for UK, as yet known only from a single locality in Wales. Though typically on *M. mantica*, the occurrence of this species on *M. erecta* has been previously reported from Bulgaria (Denchev 1997, as *Bauhinus jehudanus*).

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This research received support from the SYNTHESYS Project (<http://www.synthesys.info/>), which is financed by European Community Research Infrastructure Action under the FP7 Integrating Activities Programme. The authors also gratefully acknowledge Dr Kálmán Vánky (Herbarium *Ustilaginales* Vánky, Tübingen, Germany) and Dr Roger G. Shivas (Queensland Primary Industries and Fisheries, Australia) for critically reading the manuscript and serving as pre-submission reviewers.

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**South Florida microfungi:  
*Kalamarospora multiflagellata* gen. et sp. nov. (hyphomycetes),  
with additional new records from USA**

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**Abstract**—*Kalamarospora multiflagellata* anam. gen. et sp. nov. is described and illustrated from rachides of dead leaves of *Sabal palmetto* collected in southwestern Florida, USA. The genus is characterized by having obclavate to ellipsoidal conidia internally filled with a mass of subhyaline, septate, 2–3 µm wide filaments growing upward from suprabasal cells at the bottom of the conidia and protruding apically or subapically as long, filiform, subhyaline or hyaline, sometimes 1–2 times dichotomously branched appendages. Conidia are borne on monoblastic, transversely striate, percurrently proliferating conidiogenous cells disposed on macronematous, cylindrical, solitary, unbranched, dark brown to blackish brown conidiophores. The conidial secession is rhexolytic, leaving a distinct, usually truncate frill up to 7 µm long, which remains attached to the basal cell of the conidia. *Kalamarospora* is compared with anamorphic genera and species having a similar internal conidial organization or morphologically close taxa with appendiculate conidia. *Ellisembia britannica*, *Polytretophora calcarata*, *Pseudoacrodictys corniculata*, *Sporidesmiella sinensis*, and *Tripodsporium verruculosum* are newly recorded from USA.

**Key words**—*Ceratosporella*, *Megacapitula*, palm fungi, *Piricaudilium*

## Introduction

During a short visit to southwestern Florida, specifically the city of Naples and surrounding areas, some plant debris was collected in order to study the associated saprobic hyphomycetes (anamorphic fungi). A conspicuous and apparently undescribed anamorph was found growing on rachides of dead leaves of *Sabal palmetto*. The fungus shows close similarities to the monotypic genus *Megacapitula* J.L. Chen & Tzean (Chen & Tzean 1993) in conidial morphology and the presence of multiple apical, filiform appendages. Upon closer examination, however, the conidia revealed a peculiar internal

structure originating the appendages in combination with other features such as macronematous conidiophores, percurrent proliferating conidiogenous cells and a rhexolytic conidial secession. These features are significantly different from *Megacapitula* as presently conceived, and to my knowledge the combination of characters exhibited by the present fungus is distinct enough from all other previously known anamorphic genera to warrant the proposal of a new genus to accommodate it. *Kalamarospora* is therefore introduced, and a new species *K. multiflagellata* is described and illustrated herein. The type specimen and semi-permanent slides are deposited in the Herbarium of the U.S. National Fungus Collections (BPI). Five other hyphomycete species are recorded for the first time from USA, including comments on their taxonomy, morphology, and geographical distribution.

## Taxonomy

### *Kalamarospora* G. Delgado, anam. gen. nov.

MYCOBANK MB518541

*Ad fungus anamorphicos, hyphomycetes, pertinens. COLONIAE in substrato naturali effusae, pilosae. MYCELIUM plerumque in substrato immersum, ex hyphis ramosis, septatis, laevibus, pallide brunneis vel brunneis compositum. STROMATA absentia. CONIDIOPHORA macronematosa, mononematosa, singula vel aggregata, simplicia, erecta, recta vel leviter flexuosa, plerumque transversaliter striata, cylindrica, septata, atrobrunnea vel nigro-brunnea, percurrenter proliferantia. CELLULAE CONIDIOGENAE monoblasticae, in conidiophoris incorporatae, terminales, cylindricae, pallide brunneae vel brunneae, transversaliter striatae, percurrentes. CONIDIORUM SECESSIO rhexolytica. CONIDIA acrogena, solitaria, obclavata vel ellipsoidea, pallide brunnea vel brunnea, laevia, massa interna hypharum subhyalinarum, septatarum impleta, hyphis compluribus sursum protrudentibus velut appendicibus filiformibus, subhyalinis vel hyalinis, nonnumquam dichotomis. TELEOMORPHOSIS ignota.*

**Species typica**—*Kalamarospora multiflagellata* G. Delgado

**ETYMOLOGY**—Greek, καλαμάρι, squid and σπόρος, seed, in reference to the squid-like shape of the conidia.

Anamorphic fungi, hyphomycetes. COLONIES on natural substratum effuse, hairy. MYCELIUM predominantly immersed in the substrate, composed of branched, septate, smooth, pale brown to brown hyphae. STROMATA none. CONIDIOPHORES macronematous, mononematous, single or in groups, simple, erect, straight or slightly flexuous, mostly transversally striate, cylindrical, septate, dark brown or blackish brown, regenerating percurrently. CONIDIOGENOUS CELLS monoblastic, integrated, terminal, cylindrical, light brown to brown, transversely striate, percurrent. CONIDIAL SECESSION rhexolytic. CONIDIA acrogenous, solitary, obclavate or ellipsoidal, light brown to brown, smooth, internally filled with a visible mass of subhyaline, septate filaments protruding apically or subapically as multiple long, filiform, subhyaline or hyaline, dichotomously branched appendages. TELEOMORPH unknown.



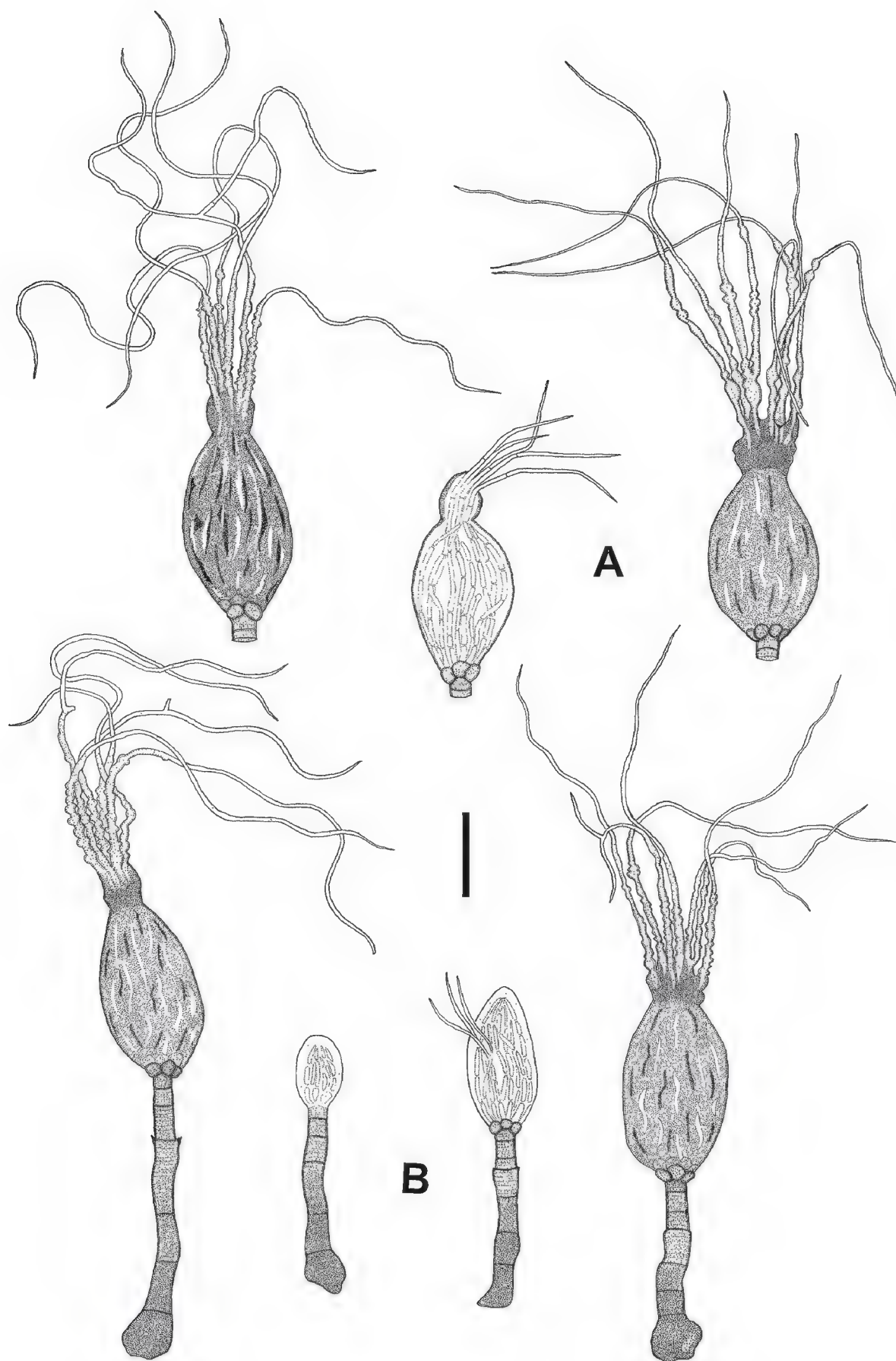


FIG. 1. *Kalamarospora multiflagellata*, from holotype (BPI 879811A).  
 A. Conidia. B. Conidiophores, conidiogenous cells and conidia.  
 The younger conidia show internal structure. Scale bar: 30  $\mu$ m.

***Kalamarospora multiflagellata*** G. Delgado, anam. sp. nov.

FIGS. 1–13

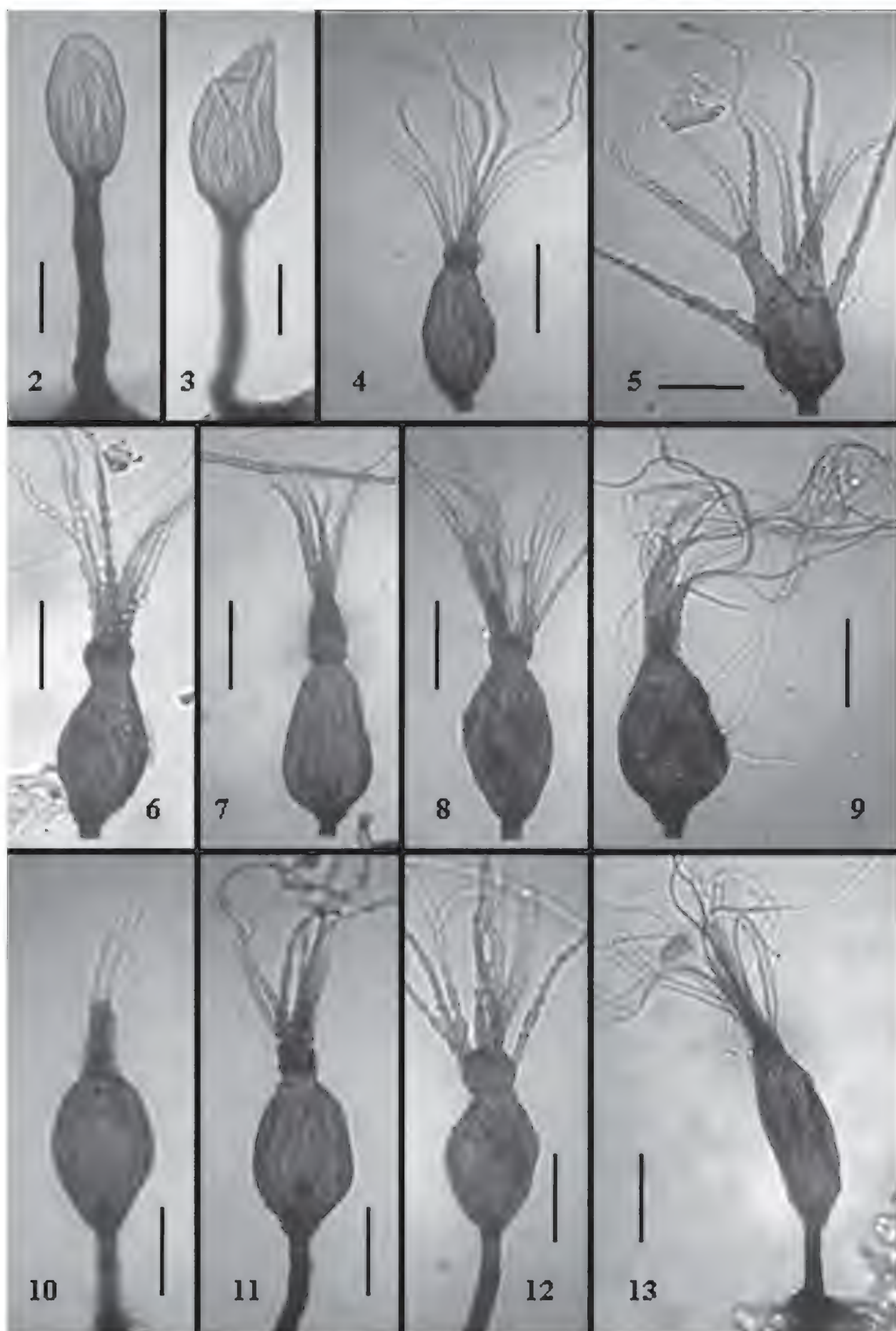
MYCOBANK MB518542

*COLONIAE* in substrato naturali effusae, brunneae, pilosae. *MYCELIUM* plerumque in substrato immersum, ex hyphis ramosis, septatis, laevibus, pallide brunneis vel brunneis, 1–2.5  $\mu\text{m}$  diam. compositum. *STROMATA* absentia. *CONIDIOPHORA* macronemata, mononemata, singula vel 2–4 aggregata, simplicia, erecta, recta vel leviter flexuosa, transversaliter striata, irregulariter verruculosa vel laevia ad basim, crassitunicata, cylindrica, septata, atrobrunnea vel nigro-brunnea, usque ad 115  $\mu\text{m}$  longa, 6–8  $\mu\text{m}$  crassa, ad basim inflata, 7–15  $\mu\text{m}$  crassa, semel ad quarter percurrenter proliferentia. *CELLULAE CONIDIOGENAE* monoblasticae, in conidiophoris incorporatae, terminales, cylindricae, pallide brunneae vel brunneae, transversaliter striatae, percurrentes, ad apicem truncatae. *CONIDIORUM SECESSIO* rhexolytica. *CONIDIA* acrogena, solitaria, obclavata vel ellipsoidea, pallide brunnea vel brunnea, tenuitunicata, laevia, 56–90  $\times$  25–45  $\mu\text{m}$  (appendice exclusa), e cellula basali, 4–6 cellulis suprabasalibus, coprore compacto fusiformi et usque ad 12 appendicibus apicalibus filiformibus composita; cellula basalis cylindrica, truncata, pallide brunnea vel brunnea, transversaliter striata, 5–8  $\times$  5–7  $\mu\text{m}$ , ad basim residuum conspicuum praebens, usque 7  $\mu\text{m}$  longa; cellulae suprabasales verticillatae, brunneae, laeves, 5–9  $\times$  4–6  $\mu\text{m}$ ; corpus conidiale massa interna filamentorum subhyalinorum, septatorum, 2–3  $\mu\text{m}$  latorum impleta, hyphis ascendentibus in summon conidio velut appendices filiformes, longae, septatae, subhyalinae vel hyalinae exeuntes, nonnumquam semel vel bis dichotomae; conidia usque ad 525  $\mu\text{m}$  longa, corpus sursum attenuatum; in parte apicali saepe inflatum, atrum, tunica mucosa conspicua, pallide brunnea vel brunnea circumdatum. *TELEOMORPHOSIS* ignota.

**HOLOTYPE**—UNITED STATES. Florida: Collier Co., NAPLES, on rachides of dead leaves of *Sabal palmetto* (Walter) Lodd. ex Schult., (*Arecaceae*), XI.23.2007, coll. G. Delgado (BPI 879811A).

**ETYMOLOGY**—Latin, *multiflagellata*, referring to the multiple filiform appendages of the conidia.

Anamorphic fungi, hyphomycetes. **COLONIES** on natural substratum effuse, brown, hairy. **MYCELIUM** predominantly immersed in the substrate, composed of branched, septate, smooth-walled, pale brown to brown hyphae, 1–2.5  $\mu\text{m}$  wide. **STROMATA** none. **CONIDIOPHORES** macronematous, mononematous, single or sometimes aggregated in groups of 2–4, simple, erect, straight or slightly flexuous, transversely striate, irregularly verruculose or smooth toward the base, thick-walled, cylindrical, septate, dark brown or blackish brown, up to 115  $\mu\text{m}$  long, 6–8  $\mu\text{m}$  wide, 7–15  $\mu\text{m}$  wide at the swollen base, with up to four successive, regenerative percurrent proliferations. **CONIDIOGENOUS CELLS** monoblastic, integrated, terminal, cylindrical, light brown to brown, transversely striate, percurrent, truncate at the apex. **CONIDIAL SECESSION** rhexolytic. **CONIDIA** acrogenous, solitary, obclavate or ellipsoidal, light brown to brown, thin-walled, smooth, often with wrinkled walls, 56–90  $\times$  25–45  $\mu\text{m}$  (excluding filaments), composed of a basal cell, 4–6 suprabasal cells, an ellipsoidal or obclavate main body, and up to 12 apical filiform appendages; basal cell cylindrical, truncate, light brown to brown, transversely striate, 5–8  $\times$  5–7  $\mu\text{m}$ , with a distinct, usually truncate, rarely irregular basal frill, up to 7  $\mu\text{m}$



FIGS. 2-13. *Kalamarospora multiflagellata*, from holotype (BPI 879811A). 2-3. Young conidia showing incipient filaments. 4-9. Conidia. 10-13. Conidiophores, conidiogenous cells and conidia. Scale bars: 2-3 = 15 µm; 4-13 = 30 µm.

long; suprabasal cells disposed side by side around the upper part of the basal cell, brown, smooth,  $5\text{--}9 \times 4\text{--}6 \mu\text{m}$ ; conidial body internally filled with a visible mass of subhyaline, septate,  $2\text{--}3 \mu\text{m}$  wide filaments, growing upward from the inner portions of the suprabasal cells at the bottom of the conidia, elongating and protruding apically or subapically as divergent filiform, septate, subhyaline or hyaline, sometimes 1–2 times dichotomously branched appendages, up to  $525 \mu\text{m}$  long, tapering to  $1 \mu\text{m}$  at the apex; apical, protrusion region usually swollen, darker and surrounded by a light brown to brown, mucilaginous sheath extending to the proximal parts of the appendages. TELEOMORPH unknown.

## Discussion

*Kalamarospora* is a genus of anamorphic, dematiaceous hyphomycetes with a unique combination of conidiogenesis, internal conidial organization and morphological features. The conidia are obclavate or ellipsoidal in shape, with thin, smooth, light brown to brown, often wrinkled walls, especially in well developed, older conidia, probably as a consequence of desiccation. They are internally filled with a visible mass of subhyaline, septate,  $2\text{--}3 \mu\text{m}$  wide filaments, which arise from the inner parts of 4 to 6 brown suprabasal cells disposed side by side around the upper portion of a cylindrical, transversely striate, light brown to brown, truncate basal cell. The inner filaments grow upward, elongating and filling the inner space of the conidium, often with terminal cells slightly swollen and rounded. They protrude more or less synchronously through the conidial apex as a bundle of long, filiform, septate, subhyaline or hyaline, divergent, 1–2 times dichotomously branched appendages. The apical area around the protrusion is usually darker and surrounded by a light brown to brown, mucilaginous sheath, often giving a swollen appearance to the apex. Occasionally, the filaments also protrude subapically, not as a bundle but individually or in groups of 2–3 filaments. Once the filaments elongate outside the conidial wall, the mucilage extends and remains surrounding the proximal parts of the appendages, showing several discontinuities, gaps or bubbles once dry. The overall conidial morphology recalls the aspect of a minute squid, hence the name of the fungus *Kalamarospora*.

The conidia are born monoblastically on cylindrical, percurrent and transversely striate conidiogenous cells. This peculiar wall ornamentation is present also in the conidial basal cells and the upper conidiophore cells, and is apparently related with the rhexolytic break of the wall of the subtending cell of the conidium. A less pigmented, annular dehiscence zone is discernible below the basal cell delimiting septum. The transverse striations may serve as dehiscence lines where the circumscissile fracture of the lateral walls is more likely to evenly occur, usually a short distance below the basal cell delimiting septum and within the dehiscence zone. As a result, the conidiogenous cell



becomes empty and open-ended, and the detached conidium bears a cylindrical, truncate, and striate frill up to 7  $\mu\text{m}$  long, which remains attached to the basal cell of the conidia. Conidiophore proliferation and subsequent conidiogenous cell delimitation occur then similarly as described for *Endophragmiella* B. Sutton (Holubová-Jechová 1986, Hughes 1979) and *Rhexoacrodictys* W.A. Baker & Morgan-Jones (Baker et al. 2002). Up to four successive, percurrent, and striate in appearance proliferations were seen in a single conidiophore, sometimes with a dark remnant of wall at the apex. However, after a percurrent proliferation emerges through the empty, non viable conidiogenous cell, two secondary septa are apparently laid down on the new proliferation, one delimiting the new conidiogenous cell and the other delimiting the basal cell of the next detached conidium. Baker et al. (2002) noticed a similar septation pattern following regenerative growth in *R. erecta*. Consequently, the conidium basal cell is more or less already established at the early stages of conidium development, showing already transversely striations. Two or three short, incipient filaments are recognizable within the conidium initial, sometimes with a very thin septum in one of them. Suprabasal cells likely originate from the lower cell of each of these incipient filaments.

Among the genera of anamorphic fungi hitherto known, the monotypic genus *Megacapitula* (Chen & Tzean 1993) closely resembles *Kalamarospora* in conidial morphology. *Megacapitula villosa* also possesses obclavate or ellipsoidal, pigmented conidia crowned with several densely packed, hairy, branched or unbranched, septate, apical appendages up to 556  $\mu\text{m}$  long. The original description did not mention an existing internal structure originating the appendages, not even in early stages of conidial ontogeny, but the apical outer wall cracks open at a certain point of conidial development, apparently peeling-off easily, and the filiform appendages emerge from the conidial apex (Chen & Tzean 1993). Unfortunately, I was unable to examine the type material to confirm the presence of such an internal structure, probably present and overlooked as a result of the opaque, dark brown or black outer conidial wall. However, *Megacapitula* and *Kalamarospora* are not considered congeneric here because they differ in certain essential features. *Megacapitula* has micronematous or semi-macronematous, simple or branched, smooth, roughened, or verrucose conidiophores, with determinate, not percurrent, terminal but also lateral or occasionally intercalary conidiogenous cells. Conidia are muriform when mature, often with a reticulate wall when young, and secede schizolytically. The apical, long, filiform appendages present in both fungi, in addition to the mucilaginous sheath surrounding the conidial apex in *Kalamarospora*, are a rare combination of features among hyphomycetes. They are probably involved in the secure attachment of the conidia to the substrate after release and dispersal (Jones 2006).



The genus *Piricaudilium* Hol.-Jech. (Holubová-Jechová 1988) possesses an internal conidial organization more or less similar to *Kalamarospora*. They both share in common the presence of conidia with an internal mass of hyaline and septate filaments arising from the inner surface of the basal part of the conidia and filling their internal space once enlarging. The two genera, however, considerably differ in conidiophore, conidial morphology, and conidiogenesis. Conidiophores in *Piricaudilium* are micronematous or semi-macronematous, sometimes consisting only of monotretic, spherical or subspherical, terminal or intercalary conidiogenous cells, with an apical pore surrounded by a distinct dark scar. The conidia are turbinate or irregular in shape, ranging from obconical, spherical or subspherical to ovoid, finely rough, verrucose to spinulose around the base, with up to 10 pale brown, thick-walled, slightly flexuous or curved setiform appendages arising from distinct lobes and up to 120  $\mu\text{m}$  long. The internal filaments are branched, apparently forming a network, and do not protrude outwards the conidial wall as in *Kalamarospora*, but instead end in short superficial appendages or fill the inner space of the longer setiform appendages. Holubová-Jechová (1988) noted that filaments in *Piricaudilium* were visible after long-term exposure to lactophenol cotton blue stain but were not colored in cotton blue. In *Kalamarospora*, however, filaments were mostly visible in younger, thinner-walled, developing conidia but also in older, even moderately wrinkled spores, which had been exposed to stain. Holubová-Jechová also considered these inner filaments were involved in the stabilization of the conidial morphology, which apparently occurs also in *Kalamarospora*, but stated that the filaments cells probably had a reproductive character as microconidia or were part of a synanamorph, which I was unable to verify in *Kalamarospora*. Future ultrastructural studies may be necessary to clarify the origin and role of these inner filaments in both fungi.

The propagules or sclerotia of the basidiomycetous anamorph *Akenomyces* G. Arnaud ex D. Hornby (Hornby 1984, Voglmayr & Krisai-Greilhuber 1997), with a complex internal and external structure, are also superficially comparable to *Kalamarospora*. They are dark brown, ellipsoidal-lenticular or obclavate among other shapes, with a tightly interwoven mass of internal, hyaline, thin-walled, much branched hyphae, 1.8–3.7  $\mu\text{m}$  wide. The presence of hyphae bearing clamp-connections at the septa and its position within the *Basidiomycota*, however, clearly separates *Akenomyces* from *Kalamarospora*. Moreover, the sclerotia originate from sclerotial initials made up of tightly interwoven hyphae, and the walls are formed by a one-celled layer of dark, thick-walled, parallel hyphae interrupted by tubercles. They are loosely and externally enclosed by upwardly growing, curved or sinuate, hyaline hyphae densely incrustated with needle-shaped crystals. Although originally collected in a terrestrial environment, the complex sclerotial structure suggests an

adaptation of *Akenomyces* to the aero-aquatic niche (Voglmayr & Krisai-Greilhuber 1997).

Some species of *Ceratosporella* Höhn. with cheiroid conidia superficially resemble *Kalamarospora* in conidial morphology and conidiogenesis. This is one of the three types of conidial morphology currently recognized within the genus. The presence of two to sixteen branches or arms arising from a basal cell and more or less closely packed in a hand-shaped appearance characterized this group of species (Castañeda 1985, Castañeda et al. 1996b, Hughes 1952, 1971, Kuthubutheen & Nawawi 1991a, Lustrati 1980, Matsushima 1981, 1993, Sinclair et al. 1987, Wu & Zhuang, 2005, Zhang et al. 2009). *Ceratosporella disticha* Kuthub. & Nawawi, *C. compacta* R.F. Castañeda et al. and *C. flagellifera* Matsush. bear the most similarity to *K. multiflagellata*, particularly in having monoblastic, percurrent conidiogenesis and compact conidia with the apical cell of each arm forming a septate, slender appendage, surrounded by a mucilaginous sheath as in the case of *C. compacta*. They differ from *Kalamarospora*, however, in having branched conidia which schizolytically secede from the conidiogenous cells and lack an internal conidial structure.

Another group of species within the genus *Pseudoacrodictys* W.A. Baker & Morgan-Jones with appendiculate conidia (Baker & Morgan-Jones 2003, Somrithipol & Jones 2003) show also a slight resemblance with *Kalamarospora*. *Pseudoacrodictys appendiculata* (M.B. Ellis) W.A. Baker & Morgan-Jones, *P. corniculata*, *P. eickeri* (Morgan-Jones) W.A. Baker & Morgan-Jones, and *P. viridescens* (B. Sutton & Alcorn) W.A. Baker & Morgan-Jones possess conidia with a distinctly protuberant basal cell delimited by a transverse septum and somewhat hyphae-like, clustered or not, septate appendages. These appendages, however, are fewer in number and shorter in length compared with those in *Kalamarospora*, the longest reaching up to 56 µm long in *P. appendiculata*. They are not originated as a result of an internal conidial structure, and occasionally break or collapse at the thin-walled tip giving a truncate aspect. The conidia also differ in shape, ranging from subglobose to broadly pyriform, turbinate or somewhat irregularly shaped, secede schizolytically and bear numerous septa arranged in an oblique fashion. The cheiroid, ellipsoidal conidia of *P. dimorphospora* Somrith. & E.B.G. Jones (Somrithipol & Jones 2003) are reminiscent of those of *C. compacta* discussed above, and a reexamination of the type specimen might be necessary to confirm if they are conspecific.

The monotypic genus *Veracruzomyces* Mercado et al. (Mercado et al. 2002) also resembles *Kalamarospora* in having monoblastic, integrated, terminal, cylindrical, percurrent proliferating conidiogenous cells and obclavate, brown conidia similar in length, with a dark brown to black, cylindrical basal cell and a mucilaginous sheath at the apex. However, the conidia in *Veracruzomyces* are muriform, rostrate, with a paler, 1–4-septate beak, seceding schizolytically

with difficult, without apical filiform appendages or internal organization, and conidiophores often bear a lateral, pale brown, lageniform and septate protuberance which bend downwards.

### Additional new records from USA

*Ellisembia britannica* (B. Sutton) W.P. Wu, in Wu & Zhuang, Fungal Diversity

Research Series 15: 116, 2005.

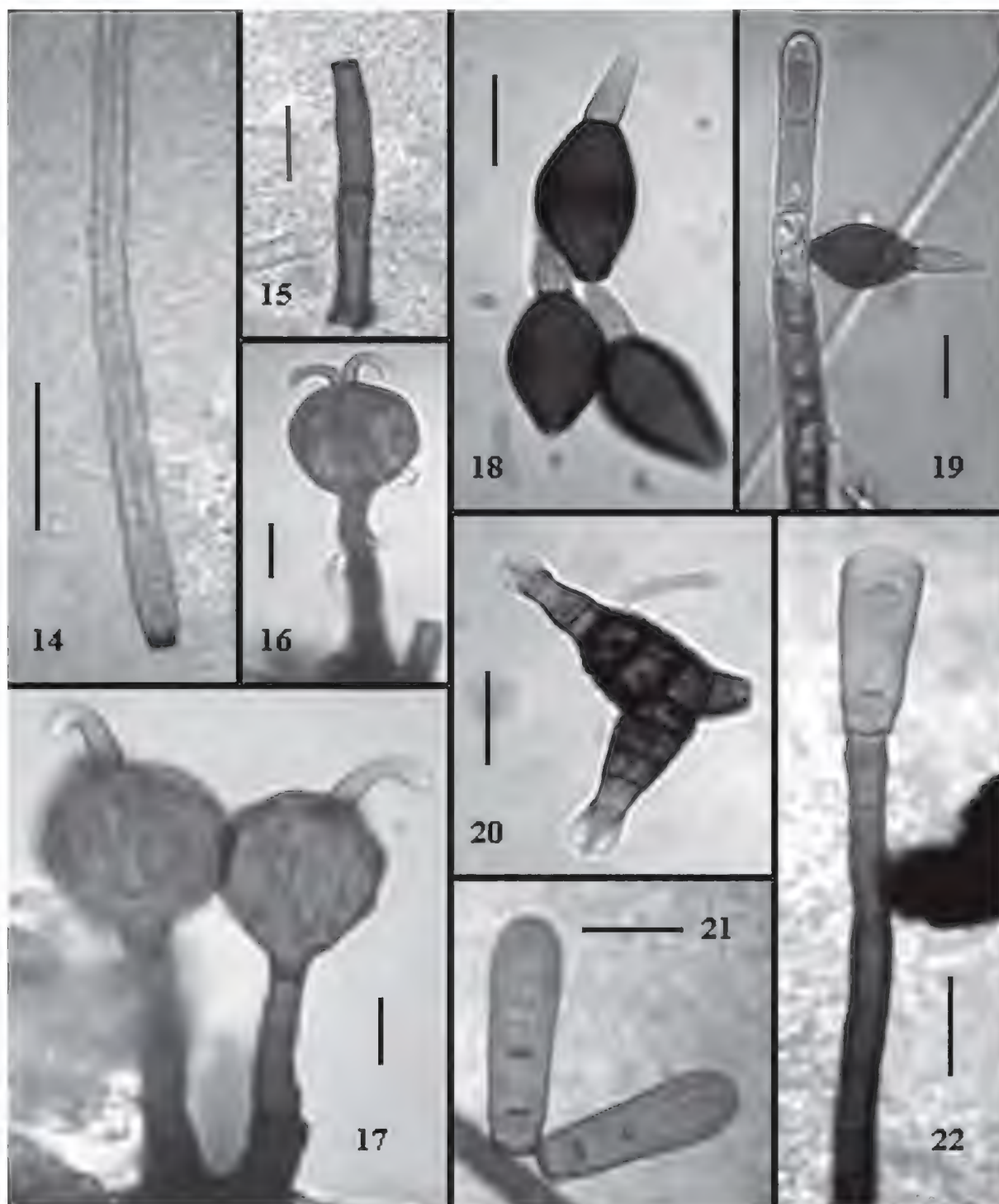
FIGS. 14–15

= *Sporidesmium britannicum* B. Sutton, in Minter, Bull. Br. mycol. Soc. 20: 87, 1986.

Colonies effuse, hairy. Conidiophores cylindrical or subcylindrical, straight or slightly flexuous, smooth, 1–3 septa, brown,  $22\text{--}47 \times 3\text{--}5\text{ }\mu\text{m}$ ; base usually bulbous, 5–6  $\mu\text{m}$  wide. Conidiogenous cells integrated, terminal, brown, apex occasionally darkened, 2–4  $\mu\text{m}$  wide. Conidia narrowly obclavate, straight to curved, 5–18-distoseptate, subhyaline to pale brown, smooth, up to 350  $\mu\text{m}$  long, 4.5–6  $\mu\text{m}$  wide; basal cell conico-truncate, darkly pigmented, 2–2.5  $\mu\text{m}$  wide at the base.

SPECIMEN EXAMINED: Florida, Collier Co., Naples, on rachides of dead leaves of *Sabal palmetto*, XI.23.2007, coll. G. Delgado (BPI 879811K).

This fungus was first described as *Sporidesmium britannicum* on a dead cupule of *Fagus sylvatica* L. from the United Kingdom (Minter 1986). Later, Wu & Zhuang (2005) collected four specimens on rotten wood and dead branches of woody plants in China, and transferred it to *Ellisembia* Subram. on the basis of its distoseptate conidia and conidiophores with irregular or without percurrent proliferations. According to the original description, conidiophores are 10–25  $\mu\text{m}$  long, often proliferate percurrently, and no mention was made of dark pigmentation in the conidial basal cells. The Florida collection is closer to the Chinese specimens in conidial features and the presence of non-proliferating conidiophores. The conidia, however, are considerably longer compared to both the holotype (up to 57.5  $\mu\text{m}$  long) and the Chinese specimens (up to 130  $\mu\text{m}$  long). Ma et al. (2008) recently described two *Ellisembia* species from China that are morphologically similar to the present specimen of *E. britannica*. *E. artocarpi* Jian Ma & X.G. Zhang and *E. sapii* Jian Ma & X.G. Zhang are characterized by very long, obclavate to long rostrate, pale brown conidia, up to 220  $\mu\text{m}$  and 240  $\mu\text{m}$  long respectively. They both differ, however, in having wider conidia without a darkened basal cell, the latter with up to 23 distosepta. Another fungus, *Sporidesmajora pennsylvaniensis* Batzer & Crous collected on fruit surface of apple in USA (Yang et al. 2010), is also comparable with the Florida specimen in having very long, narrowly obclavate to long obclavate conidia up to 350  $\mu\text{m}$  long, with darkly pigmented, obconical basal cells, but differs in its smooth to finely verruculose, guttulate and euseptate rather than distoseptate conidia.



FIGS. 14–15. *Ellisembia britannica* (BPI 879811A). 14. Conidium. 15. Conidiophore. 16–17. *Pseudoacrodictys corniculata* (BPI 880521A). Conidiophores and conidia. 18–19. *Polytretophora calcarata* (BPI 880519A). 18. Conidia. 19. Conidiophore with attached conidium. 20. *Triposporium verruculosum* (BPI 880518B). Conidium. 21–22. *Sporidesmiella sinensis* (BPI 880520A). 21. Conidia. 22. Conidiophore with attached young conidium. Scale bars: 14 = 20  $\mu$ m; 15–22 = 10  $\mu$ m.

***Polytretophora calcarata*** Mercado, Acta Bot. Cubana 16: 3, 1983. FIGS. 18–19  
 = *Spadicoides calcarata* (Mercado) Melnik, Nov. sist. Niz. Rast. 28: 68, 1992.  
 = *Parahelminthosporium malabaricum* Subram. & Bhat, Kavaka 15: 63, 1989 [“1987”].

Colonies effuse, hairy, brown. Conidiophores erect, straight or flexuous, unbranched but sometimes sparingly branched, brown, paler towards the

apex, dark brown towards the base, up to 700  $\mu\text{m}$  long, 5–7  $\mu\text{m}$  wide in the upper part, 6–11  $\mu\text{m}$  wide in the middle, 12–17  $\mu\text{m}$  wide at the base, up to 2 regenerating percurrent proliferations. Conidiogenous cells polytretic, terminal or intercalary, cylindrical, rounded at the apex when terminal. Conidia 2-celled, 24–32  $\mu\text{m}$  long; basal cell ellipsoidal to fusiform, brown, thick-walled, guttulate, often with a slightly darker band around the middle,  $13\text{--}20 \times 7\text{--}11 \mu\text{m}$ , truncate at base; apical cell subhyaline, conico-truncate, 8–13  $\mu\text{m}$  long, 3–4  $\mu\text{m}$  wide at the base, tapering to 2  $\mu\text{m}$  at the apex.

SPECIMEN EXAMINED: Florida, Collier Co., Naples, on segments of dead leaves of *Sabal palmetto*, XI.24.2007, coll. G. Delgado (BPI 880519A).

*Polytretophora calcarata*, the type species of the genus, is apparently pantropical in distribution. The fungus has been widely collected on *Arecaceae* and *Pandanaceae* in many tropical and subtropical Asian countries, Australia, and the Pacific Islands, as well as the Seychelles (Kuthubutheen & Nawawi 1991b, Whitton et al. 2001). In the Americas, it has been previously recorded several times from Cuba, the type locality (Mercado 1983, Hernández & Mena 1995, Mercado et al. 1997), on decaying palm petioles from Peru (Matsushima 1993) and now for the first time from the subtropical United States. The Florida specimen has occasionally branched, longer conidiophores compared with the holotype from Cuba (conidiophores simple, 150–350  $\mu\text{m}$  long), but is similar in conidiophore length, branching, and conidial dimensions to other specimens cited in the literature. The presence of a darker band of pigmentation around the middle of the basal cells of the conidia was originally reported by Whitton et al. (2001) and was detected in the present specimen. Kuthubutheen & Nawawi (1991b) also reported a *Selenosporella* synanamorph in collections from Malaysia, but this feature was not observed.

*Pseudoacrodictys corniculata* (R.F. Castañeda) W.A. Baker & Morgan-Jones, Mycotaxon 85: 378, 2003.

FIGS. 16–17

= *Acrodictys corniculata* R.F. Castañeda, Deuteromycotina de Cuba, Hyphomycetes 2: 1, 1985.

Colonies effuse, hairy. Conidiophores solitary or in small groups, mostly unbranched or sparingly branched, cylindrical, straight or slightly flexuous, smooth, brown to dark brown,  $23\text{--}52 \times 3\text{--}5 \mu\text{m}$ , 6–8  $\mu\text{m}$  wide at base, with 0–2 percurrent proliferations. Conidiogenous cells monoblastic, integrated, terminal, cylindrical, percurrent. Conidia subglobose to globose, rarely broadly pyriform, dictyoseptate, smooth, brown,  $17\text{--}28 \times 14\text{--}30 \mu\text{m}$ , with a distinct protuberant, conico-truncate basal cell,  $3\text{--}6 \times 4\text{--}6 \mu\text{m}$ , and 0–6 pale brown, horn-like, strongly curved, aseptate appendages, clustered or not,  $6\text{--}19 \times 2\text{--}4 \mu\text{m}$ . Conidial secession schizolytic.

SPECIMEN EXAMINED: Florida, Collier Co., Naples, on rachides of dead leaves of *Sabal palmetto*, XI.23.2007, coll. G. Delgado (BPI 880521A).



Castañeda (1985) originally described this peculiar anamorph as *Acrodictys corniculata* from fallen leaves of unidentified *Poaceae* in Cuba. Later, Baker & Morgan-Jones (2003) partly reformatted and amended the original description to accommodate it, along with six other formerly placed *Acrodictys* species, within the narrowly delimited genus *Pseudoacrodictys*. *Pseudoacrodictys corniculata* is distinct by the presence of relatively small conidia, with short, horn-like, strongly curved appendages, often distally clustered at the apex. The present collection is the second record of its occurrence worldwide. The Florida specimen is similar to the holotype in dimensions and morphology, but sometimes the conidial appendages were not apically clustered but segregated and laterally placed, especially in larger, broadly pyriform conidia. Conidiophores are occasionally branched, often showing an irregular tear of the proximal periclinal wall, and conidia sometimes carried away a more or less short piece of conidiophore once released, a feature mentioned by Baker & Morgan-Jones (2003) and not related with rhexolytic secession.

*Sporidesmiella sinensis* W.P. Wu, in Wu & Zhuang,  
Fungal Diversity Research Series 15: 176, 2005.

FIGS. 21–22

Colonies effuse, brown, hairy. Conidiophores cylindrical, straight or flexuous, smooth, brown, paler toward the apex, up to 131  $\mu\text{m}$  long, 3–5  $\mu\text{m}$  wide, 6–10  $\mu\text{m}$  wide at base, with up to 7 inconspicuous annellidic percurrent proliferations. Conidia clavate, 3-distoseptate, rarely 2 or 4, cell lumina reduced, pale olivaceous to pale brown, 18–26  $\times$  5–7.5  $\mu\text{m}$ ; apex rounded, basal cell slightly darker, truncate, 4  $\mu\text{m}$  wide at the base.

SPECIMEN EXAMINED: Florida, Collier Co., Naples, on dead liana stems, XI.24.2007, coll. G. Delgado (BPI 880520A).

*Sporidesmiella sinensis* was recently described from dead twigs in China (Wu & Zhuang 2005). The original discussion did not include *S. oraniopsis* Yanna et al., a morphologically similar species having also percurrent proliferating conidiophores and 3-distoseptate, pale-colored, rounded at the apex, truncate at the base, clavate conidia (Yanna et al. 2001). *Sporidesmiella sinensis*, however, has smaller (24–26  $\times$  7.5–9  $\mu\text{m}$ ), also cuneiform, pale olivaceous to olivaceous brown conidia and inconspicuous, 4–8 annellidic proliferations, while *S. oraniopsis* has pale brown, larger conidia (28–40  $\times$  8–10  $\mu\text{m}$ ), rarely with 4 to 5 distosepta, and conspicuous, up to 18 percurrent proliferations at the apex. The Florida specimen agrees fairly well with the holotype description of *S. sinensis*, but conidia are narrower and rarely 2 or 4-distoseptate.

*Triposporium verruculosum* R.F. Castañeda, Gené & Guarro,  
Mycotaxon 59: 207, 1996.

FIG. 20

Colonies hairy, effuse. Conidiophores cylindrical, straight or slightly flexuous, smooth, brown, up to 100  $\mu\text{m}$  long, 4–6  $\mu\text{m}$  wide, basal cells dark brown, 8–10

µm wide. Conidiogenous cells monoblastic, integrated, terminal, cylindrical, occasionally with 1–2 doliiform percurrent proliferations, slightly attenuated and truncate at the apex. Conidia stauriform, composed of a brown, obconical or cylindrical basal cell, 4–8 × 4.5–6 µm, a dark brown, verrucose suprabasal cell, 4–6 × 5–8 µm, and 2–4 divergent, verruculose, brown arms, 3–5-septate, 14–28 µm long, 7–9 µm wide at base, paler toward the apex and frequently ending in a rounded drop of mucilage, 3.5–5 µm diam.

SPECIMEN EXAMINED: Florida, Collier Co., Naples, on rachides of dead leaves of *Sabal palmetto*, XI.23.2007, coll. G. Delgado (BPI 880518B).

*Triposporium verruculosum* morphologically resembles *T. elegans* Corda the type species of the genus (Ellis 1971, Wu & Zhuang 2005), but differs in having verruculose, smaller conidial arms. The fungus was originally described on rotten fallen leaf of *Laurus* sp. from Canary Islands (Castañeda et al. 1996a). A second specimen collected on dead leaf of *Quercus ilex* L. from New Zealand is deposited in PDD (NZFUNGI 2010). The Florida collection has shorter conidiophores compared with the holotype (120–260 µm long).

### Acknowledgments

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**A new species of *Colletotrichum* from *Cordyline fruticosa* and *Eugenia javanica* causing anthracnose disease**

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**Abstract** — A new species *C. cordylinicola*, isolated from *Cordyline fruticosa*, is characterized by morphological and molecular characters. The species would previously have been considered as a member of the *Colletotrichum gloeosporioides* complex. Combined six gene analysis using ACT, GS, TUB2, ITS, CAL and GPDH shows that three strains of *C. cordylinicola* clustered in a distinct lineage as a sister clade to *C. kahawae*. Other reference taxa employed in the analysis include type strains of *C. asianum*, *C. fruticola*, *C. gloeosporioides*, *C. kahawae*, *C. siamense*, *C. simmondsii*, and authentic strains of *C. horii*. This is the first report of a *Colletotrichum* species causing disease of *Cordyline fruticosa* in Thailand. Pathogenicity testing using the strains isolated from *Cordyline fruticosa* and *Eugenia javanica* showed that two strains isolated from different hosts may represent different pathotypes.

**Key words** — leaf spot, plant pathogenic fungi, taxonomy

## Introduction

*Colletotrichum* is one of the most economically important pathogenic genera causing anthracnose of fruits and leaves, affecting a wide range of hosts in the tropics and subtropics. (Freeman et al. 1998, Hindorf 2000, Damm et al. 2009, Hyde et al. 2009a,b, Shivas & Yu 2009). Both agricultural crops and fruit trees

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TABLE 1. Sources of isolates used in this study and analysis.

Colletotrichum species	CULTURE COLLECTION	GENBANK ACCESSION NUMBER					
		ACT	TUB-2	CAL	GS	GPDH	ITS
<i>C. asianum</i>	MFU 090232*	FJ 903188	FJ 907434	FJ 917501	FJ 972586	FJ 972571	FJ 972605
<i>C. asianum</i>	MFU 090233	FJ 907424	FJ 907439	FJ 917506	FJ 972595	FJ 972576	FJ 972612
<i>C. asianum</i>	MFU 090234	FJ 907421	FJ 907436	FJ 917503	FJ 972598	FJ 972573	FJ 972615
<i>C. cordylinicola</i>	BCC 38872	HM470234	HM470249	HM470237	HM470243	HM470240	HM470246
<i>C. cordylinicola</i>	BCC38864	HM470233	HM470248	HM470236	HM470242	HM470239	HM470245
<i>C. cordylinicola</i>	MFLU 100132	HM470235	HM470250	HM470238	HM470244	HM470241	HM470247
<i>C. fructicola</i>	MFU 090226*	FJ 907427	FJ 907442	FJ 917509	FJ 972592	FJ 972579	FJ 972602
<i>C. fructicola</i>	MFU 090227	FJ 907425	FJ 907440	FJ 917507	FJ 972594	FJ 972577	FJ 972611
<i>C. fructicola</i>	MFU 090228	FJ 907426	FJ 907441	FJ 917508	FJ 972593	FJ 972578	FJ 972603
<i>C. gloeosporioides</i>	CBS 953.97*	FJ 907430	FJ 907445	FJ 917512	FJ 972589	FJ 972582	FJ 972609
<i>C. horii</i>	TSG001	GU133374	GU133375	GU133376	GU133377	GQ329682	AY787483
<i>C. horii</i>	TSG002	GU133379	GU133380	GU133381	GU133382	GQ329680	AY791890
<i>C. kahawae</i>	IMI 319418*	FJ 907432	FJ 907446	FJ 917514	FJ 972588	FJ 972583	FJ 972608
<i>C. kahawae</i>	IMI 363578*	FJ 907433	FJ 907447	FJ 917515	FJ 972587	FJ 972584	FJ 972607
<i>C. siamense</i>	MFU 090230*	FJ 907423	FJ 907438	FJ 917505	FJ 972596	FJ 972575	FJ 972613
<i>C. siamense</i>	MFU 090231	FJ 907422	FJ 907437	FJ 917504	FJ 972597	FJ 972574	FJ 972614
<i>C. simmondsii</i>	BRIP 28519*	FJ 907428	FJ 907443	FJ 917510	FJ 972591	FJ 972580	FJ 972601
<i>C. simmondsii</i>	CBS 294.67	FJ 907429	FJ 907444	FJ 917511	FJ 972590	FJ 972581	FJ 972610
<i>C. falcatum</i>	CGMCC3.14187	HM171665	HM171680	HM171668	HM171674	HM171671	HM171677

NOTE: ACT: actin; TUB-2: partial  $\beta$ -tubulin; CAL: calmodulin; GS: glutamine synthetase; GPDH: glyceraldehydes-3-phosphate dehydrogenase; ITS: complete rDNA-ITS region. The newly generated sequence in this study are shown in bold. CGMCC: China General Microbial Culture Collection. MFU: Mae Fah Luang University, Thailand; CBS: Centraalbureau voor Schimmelmcultures, Utrecht, The Netherlands; BRIP: Plant Pathology Herbarium, Department of Primary Industries, Queensland, Australia; IMI: CABI Europe – UK, Egham, Surrey TW209TY, UK; \*: ex-type cultures

can be affected by *Colletotrichum* anthracnose, resulting in reduction in yield quantity or quality. *Colletotrichum* species are cosmopolitan with either multiple species occurring on a single host or a single species on multiple hosts (Cai et al. 2009, Crouch & Beirn 2009, Hyde et al. 2009b). Fungus/host relationships are broad, imprecise and often overlapping. *Colletotrichum* species can infect many hosts and may adapt to new environments (Sanders & Korsten 2003a), leading to serious cross infection problems in plant production. The study of pathogenic variability of *Colletotrichum* species is therefore important and the understanding of the host range of a particular pathogen may help in efficient disease control and management (Whitelaw-Weckert et al. 2007).

Artificial inoculation methods in vitro are commonly used to test the pathogenicity of a fungal species, as it is easy to control environmental conditions. Common inoculation methods for pathogenicity testing include drop inoculation and wound/drop inoculation (Cai et al. 2009, Kanchanadomkan et al. 2004, Lin et al. 2002, Sharma et al. 2005, Than et al. 2008a).

*Colletotrichum gloeosporioides* sensu lato has previously been listed as causing disease of a very wide range of fruits and infecting leaves of many hosts in Thailand (and Laos) (Ratanacherdchai et al. 2007, Than et al. 2008b, Yang et al. 2009). This species has recently been epitypified with a living strain that has been sequenced with sequence data deposited in GenBank (Cannon et al. 2008). This has enabled researchers to compare their isolates of *Colletotrichum* with the *C. gloeosporioides* epitype. This has resulted in the description of several new species in the *C. gloeosporioides* species complex (Prihastuti et al. 2009, Yang et al. 2009). With the introduction of several new species it is important to establish whether they are host-specific or have a wide host range, as this will have important implication in disease control and management. The objective of this paper is to introduce a new *Colletotrichum* species causing leaf disease of *Cordyline fruticosa* in Laos and Thailand. It is characterized morphologically and phylogenetically in this paper and its ability to infect several hosts is established.

## Material and methods

### Isolation and morphological examination

The methods of isolation used by Cai et al. (2009), Prihastuti et al. (2009) and Yang et al. (2009) were followed. Two strains of *Colletotrichum* were isolated from anthracnose of infected leaves of *Cordyline fruticosa* from a garden in Chiang Mai, Thailand and one from leaves of rose apple in a garden in Vientiane, Laos. The growth rate was measured for 7-day old colonies on PDA. Herbarium material is deposited in MFLU while exotype cultures are deposited at MFLUCC and BIOTEC Culture Collection (BCC), with some duplicate strains deposited in China General Microbial Culture Collection (CGMCC) under material transfer agreement 7/2552.

### DNA extraction

Isolates were grown on PDA and incubated at 27°C for 7 days. Genomic DNA was extracted by using a Biospin Fungus Genomic DNA Extraction Kit (BioFlux<sup>®</sup>) according to the manufacturer's protocol. Quality and quantity of DNA were estimated visually by staining with ethidium bromide on 1% agarose gel electrophoresis.

### PCR amplification and DNA sequencing

Partial actin (ACT),  $\beta$ -tubulin (TUB2), calmodulin (CAL), glutamine synthetase (GS), glyceraldehyde-3-phosphate dehydrogenase (GPDH) genes and the complete rDNA-ITS (ITS) region from three strains were amplified by PCR reactions. The primers, reaction system and thermo cycles were the same as used by Prihastuti et al. (2009).

PCR products were verified by staining with ethidium bromide on 1% agarose electrophoresis. PCR products were then purified using the GFX PCR Purification Kit (27-9602-01; Amersham Biosciences) according to the manufacturer's protocol. Sequencing was carried out at the SinoGenoMax Company Limited, Beijing.

### Phylogenetic analyses

Sequences of *Colletotrichum* isolates (TABLE 1) from different hosts were aligned with ClustalX (Thompson et al. 1997) and optimized manually to allow maximum alignment and maximum sequence similarity. Gaps were treated as missing data. Phylogenetic analysis was carried out based on the aligned dataset by PAUP<sup>\*</sup> 4.0b10 (Swofford 2000). Ambiguously aligned regions were excluded from all analyses. Trees were inferred using the heuristic search option with TBR branch swapping and 1000 random sequence additions. Maxtrees were unlimited, branches of zero length were collapsed, and all multiple parsimonious trees were saved. Descriptive tree statistics such as tree length [TL], consistency index [CI], retention index [RI], rescaled consistency index [RC], homoplasy index [HI], and log likelihood [-ln L] (HKY model) were calculated for trees generated under different optimality criteria. Kishino-Hasegawa tests (Kishino & Hasegawa 1989) were performed in order to determine whether trees were significantly different. Clade stability of the tree resulting from maximum parsimony analysis was assessed by bootstrap analysis with 1000 replicates, each with 10 replicates of random stemwise addition of taxa (Felsenstein 1985). Trees were figured in TreeView (Page 1996).

The model of evolution was estimated by using Mrmodeltest 2.2 (Nylander 2004). Posterior probabilities (PP) (Rannala & Yang 1996, Zhaxybayeva & Gogarten 2002) were determined by Markov Chain Monte Carlo sampling (BMCMC) in MrBayes 3.0b4 (Huelsenbeck & Ronquist 2001). Six simultaneous Markov chains were run for 1,000,000 generations and trees were sampled every 100 generations (resulting in 10,000 total trees). The first 2000 trees, which represented the burn-in phase of the analyses, were discarded and the remaining 8000 trees were used for calculating posterior probabilities (PP) in the majority rule consensus tree.

### Pathogenicity testing

The protocol followed the methods outlined by Cai et al. (2009) and Yang et al. (2009), modified as follows. Pathogenicity testing used one isolate from *Cordyline fruticosa* and one from *Eugenia javanica*. Each was inoculated onto three fruits of chilli

(*Capsicum* sp.), guava (*Psidium guajava*), mango (*Mangifera indica*), papaya (*Carica papaya*), orange (*Citrus* sp.), and rose apple (*Eugenia javanica*) and onto three detached leaves of *Cordyline fruticosa*. Incubation duration was dependent on the nature of lesion development and anthracnose symptoms were scored as a + or –.

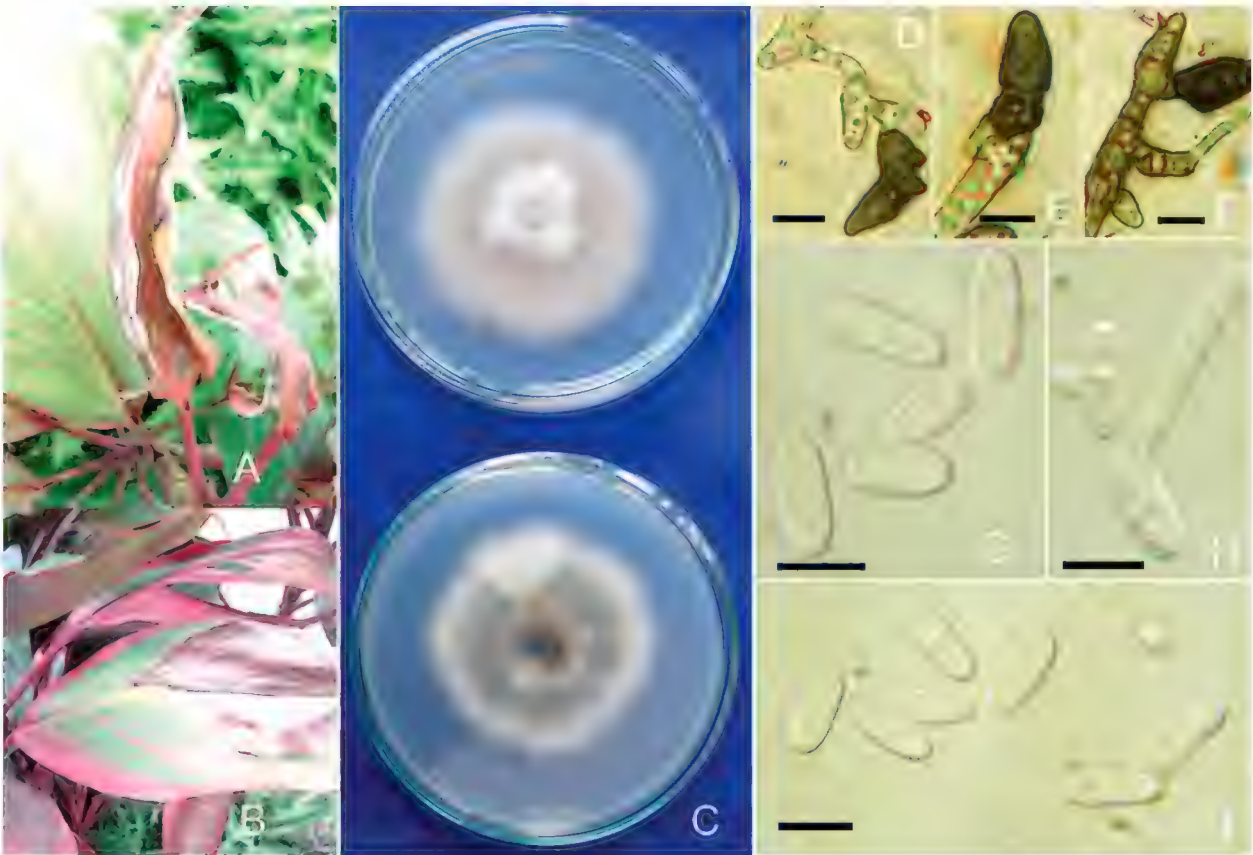


FIGURE 1. *Colletotrichum cordylinicola* (from BCC 38872, holotype) (A, B) symptoms on *Cordyline fruticosa*. (C) upper and lower view of cultures on PDA after 7 days growth; (D, E,F) appressoria; (G, I) conidia; (H) conidia germination (Bars: D–I = 10 µm).

Results

Taxonomy

*Colletotrichum cordylinicola* Phoulivong, L. Cai & K.D. Hyde, sp. nov.      FIGURE 1  
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*Coloniae crescentes post 7 dies in PDA ad 27°C 75 mm diam. Conidiogenae producentia in acervulis, tubulosa. Conidia 11–20 × 4–5 µm, unicellulares, hyalinae, cylindrici, laevia ad apicem obtuse. Appressoria 13–13.4 × 7.2–7.3 µm, brunnea vel atro-brunnea, irregulariter ovoidea vel clavati.*

**HOLOTYPE:** Thailand, Chiang Mai Province, San Sai District, Maejo Village, on *Cordyline fruticosa* (L.) A. Chev. (Agavaceae), 15 March 2009, Sitthisack Phoulivong, MFLU10 0289; extype living culture MFUCC 090551, BCC 38872 and CGMCC.

**ETYMOLOGY:** Referring to the host genus *Cordyline*.

Colonies on PDA attaining 75 mm diam. in 7 days at 27°C, growth rate 10.8–11.6 mm/day (mean= 11.2, n = 5), white, sparse, with grey-orange visible conidial masses and with floccose aerial mycelia in centre, reverse slightly greenish.



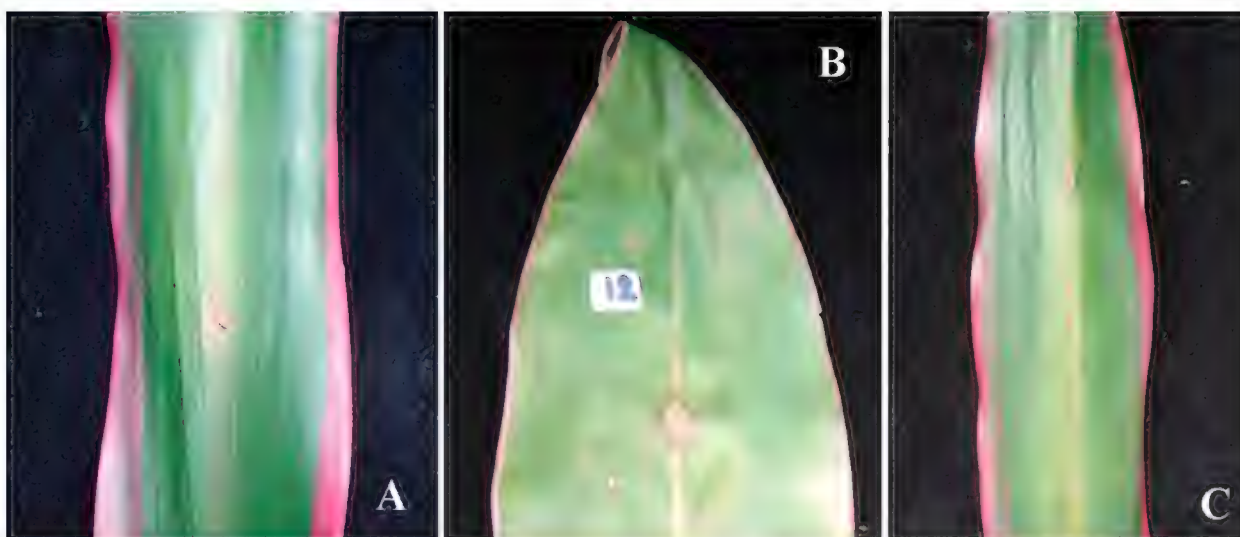


FIGURE 2. Anthracnose symptom on *Cordyline fruticosa* 7 days after inoculation.

Sclerotia absent. Setae absent. Conidiophores congregative, straight or geniculate, produced in the acervuli. Conidia  $11\text{--}20 \times 4\text{--}5 \mu\text{m}$  (mean =  $15.37 \pm 0.6 \times 4.5 \pm 0.56$ ,  $n = 30$ ), one-celled, hyaline, cylindrical with round ends, smooth-walled, guttulate. Spore germination on PDA mostly observed near the apex of the conidia, sometimes from the centre. Appressoria in slide culture  $13\text{--}13.4 \times 7.2\text{--}7.3 \mu\text{m}$  (mean =  $13.20 \pm 0.94 \times 7.25 \pm 0.61$ ,  $n = 10$ ), mostly formed from conidia, brown to dark brown, variable in shape including ovoid, clavate or slightly irregular, often becoming complex with age.

TELEOMORPH — not produced in culture.

KNOWN DISTRIBUTION — Thailand and Laos.

ADDITIONAL SPECIMENS EXAMINED: LAOS, Vientiane, Pakngum District, Don-ngaeng Village, on rose apple fruit (*Eugenia javanica* Lam. (Myrtaceae)), 26 September 2007, Sitthisack Phoulivong, MFLU10 0290, living culture MFLU 09 0636, IFRD 2149, BCC38864, CGMCC 3.14199. THAILAND, Chiang Rai, Doi Tung, on *Cordyline fruticosa*, Noireung Parinn, MFLU10 0291, living cultures MFLU 100132, CGMCC 3.14200.

### Phylogenetic study

The dataset of six combined genes comprised 2506 characters after alignment, of which 545 characters were parsimony informative (21.7%). The KH test showed that the two trees inferred from parsimonious analysis were not significantly different. One of the most parsimonious trees (TL = 1377, CI = 0.895, RI = 0.881, RC = 0.798, HI = 0.105) generated from dataset of six combined gene regions is shown in FIGURE 3 The phylograms inferred from single genes ACT, GS, TUB2, ITS, CAL and GPDH show similar topology as that from combined datasets but with much lower statistical support for branches (results not shown). In the phylogenetic tree, three strains of *C. cordylinicola* clustered in a distinct lineage and appeared as a sister clade to *C. kahawae* (100% bootstrap and posterior probability). Other reference taxa employed in the analysis include type strains



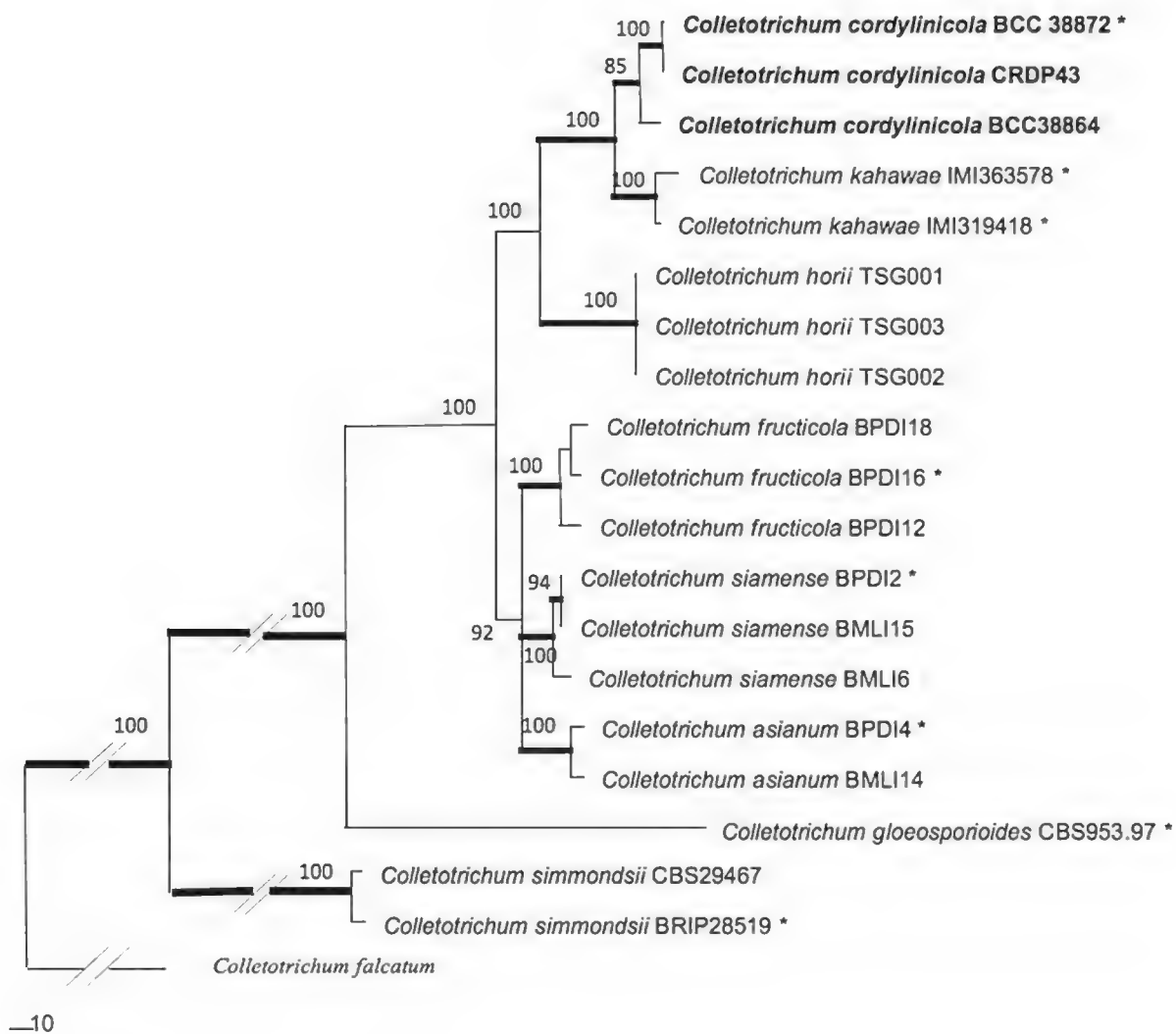


FIGURE 3. Maximum parsimony phylogram showing phylogenetic relationships among isolates of *Colletotrichum cordylinicola* and closely related taxa based on combined ACT, TUB2, CAL, GS, ITS, and GPDH sequences. Data were analysed with random addition sequence, unweighted parsimony, and treating gaps as missing data. Values above the branches are parsimony bootstrap (equal or above 50%). Thickened branches represent significant Bayesian posterior probabilities (equal or above 95%). The tree is rooted with *Colletotrichum falcatum*. \* indicates sequences from type specimens.

of *C. simmondsii*, *C. asianum*, *C. fructicola*, *C. gloeosporioides*, *C. kahawae*, *C. siamense* and authentic strains of *C. horii*.

**Pathogenicity testing**

Two isolates of *C. cordylinicola* were tested for their pathogenicity and potential for cross infection. In inoculation tests, the strain isolated from *Cordyline fruticosa* infected *Cordyline fruticosa* leaves (FIGURE 2.) and papaya fruit but did not infect the other fruits tested. The *C. cordylinicola* isolate from rose apple infected rose apple as well as citrus, chilli, guava, mango and papaya fruits but not *Cordyline fruticosa* leaves. The qualitative comparison of symptom development on different hosts is shown in TABLE 2.

TABLE 2: Pathogenicity and potential of cross infection of *Colletotrichum cordylinicola* on a range of hosts.

ISOLATE NUMBER	HOSTS	Infection on inoculated fruits*						Infection on inoculated leaves of <i>C. fruticosa</i>
		CHILLI	GUAVA	MANGO	ORANGE	PAPAYA	ROSE APPLE	
BCC 38872	<i>Cordyline fruticosa</i>	-	-	-	-	+	-	+
BCC 38864	<i>Eugenia javanica</i>	+	+	+	-	+	+	-

Discussion

*Colletotrichum cordylines* Pollacci (from Italy) is the only species of *Colletotrichum* described from *Cordyline* (*C. indivisa*). Conidial sizes were not provided in the protologue (Pollacci 1899: 44; Saccardo & Sydow 1899: 1017) and the name has not recently been used (Hyde et al. 2009b). It is impossible to establish whether our collections have any relationship to the type of *C. cordylines*, as there are no living extype cultures and it is presently impossible to isolate DNA from such an old type specimen. It is, therefore, prudent to introduce our collections as a new species.

*Colletotrichum cordylinicola* is morphologically similar to several species in the *C. gloeosporioides* complex. Species in this complex are difficult to differentiate based solely on morphology. Phylogenetic analysis using ITS sequences could not confidently resolve its systematic placement but showed that this fungus is well clustered in the *C. gloeosporioides* complex (details not shown). A multi-locus phylogeny based polyphasic approach was therefore employed to infer interspecific relationships in this group of fungi (Cai et al. 2009). In the six-gene combined phylogeny, the species relationships are well defined with all the major clades supported by parsimony bootstrap support and Bayesian posterior probabilities (FIGURE 3). The conidial morphology of *C. cordylinicola* is similar to that of *C. siamense*. However, *C. cordylinicola* can be distinguished from this species by its appressoria, which are irregular in shape (FIGURE 1). In the phylogenetic tree, *C. cordylinicola* does not group with *C. siamense*, but clusters as a sister clade to *C. kahawae* (FIGURE 3). Although similar in conidial morphology, *C. cordylinicola* can be differentiated from *C. kahawae* by its significantly larger appressoria ( $13\text{--}13.4 \times 7.2\text{--}7.3$  vs  $4.5\text{--}10 \times 4\text{--}7 \mu\text{m}$ ) and smaller conidia. This is the first report of *Colletotrichum* species causing anthracnose on *Cordyline fruticosa* in Thailand.

Identification of species within the *C. gloeosporioides* complex has been a difficult issue as these species are morphologically very similar (Bailey & Jeger 1992, Sutton 1992). Morphology of conidia and appressoria, colony characters, host association, growth rate, and biochemical data should be used in conjunction

with a multilocus phylogeny to identify a *Colletotrichum* species accurately (Cai et al. 2009; Prihastuti et al. 2009). In this study, a phylogenetically well-defined lineage is associated with distinct morphological and other phenotypic characters. It is therefore given species rank and described as a new species.

The strain of *C. cordylinicola* isolated from rose apple failed to infect *Cordyline fruticosa*, while that from *Cordyline fruticosa* failed to infect rose apple. In morphology, the two strains are essentially similar except the one from rose apple produced conidia that are slightly acute at one end, while the conidia in the strain from *Cordyline fruticosa* are rounded at both ends. The strains are, however, shown to be related based on multigene phylogenetic analysis with 100% support (FIGURE 3). The strain from rose apple infected more fruits than that from *Cordyline fruticosa*. This finding supports the statement of Johnston (2000) that “there are no general rules concerning host relationships within *Colletotrichum* . . . the group so recognized cannot be assumed genetically equivalent, even when appearing to be biologically similar”. It will be interesting to establish whether these strains represent two pathotypes in nature (Bailey & Jeger 1992). Pathogenicity may be affected by several environmental factors such as variety and condition of the fruit, humidity and temperature, and the concentration of inoculum (Simmonds 1965, Freeman et al. 1998). The result reported here may not accurately reflect the true virulence potential. Future research should attempt to determine the pathogenicity of these strains according to natural infections rather than artificial inoculations. On the other hand, if more phenotypic divergence of these two strains could be identified following further collections or study, the systematic relationship between the two strains may need a re-evaluation.

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## MYCOTAXON

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**Taxonomic studies of *Dactylella* from Fujian, China**

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**Abstract** —A new species of *Dactylella* was found during a continuing survey of anamorphic fungi in tropical areas of Fujian province, China. The new species, *Dactylella yoaniae* was found on *Yoania japonica*. It is described, illustrated and compared with closely related taxa.

**Key words** — hyphomycetes, taxonomy

**Introduction**

The genus *Dactylella* was established by Grove (1884) with *D. minuta* Grove as the type species. *Dactylella* is characterized as “Saprophytic. Vegetative hyphae creeping, sparse. Conidiophores erect, simple, septate or non-septate, smooth, hyaline. Conidia born singly at the apex of conidiophore, ellipsoidal or fusoid or cylindrical, one-celled at first, later 2- to many-septate, hyaline”. These characters separate *Dactylella* from several similar genera, viz. *Arthrobotrys* Corda, *Dactylaria* Sacc., *Monacrosporium* Oudem., *Brachyphoris* Juan Chen et al., *Drechsleromyces* Subram., *Monacrosporiella* Subram., *Gangliophragma* Subram., and *Lactydina* Subram. (Subramanian 1963, 1977; Chen et al. 2007a).

*Dactylella* is extremely heterogeneous, and many species are predatory on microanimals. Some are oospore or nematode-egg parasites while others are saprobic on deciduous stems or wood (Chen et al. 2007b). Worldwide, more than 100 species have been validly described, of which 28 species have been described from China.

Fungi were collected on dead branches or rotten wood from tropical forest in Fujian province of China during 2009. Among the collections an undescribed species of *Dactylella* was found. The type specimen is deposited in HSAUP (Herbarium of the Department of Plant Pathology, Shandong Agricultural University) with isotype in HMAS (Mycological Herbarium, Institute of Microbiology, Chinese Academy of Sciences).

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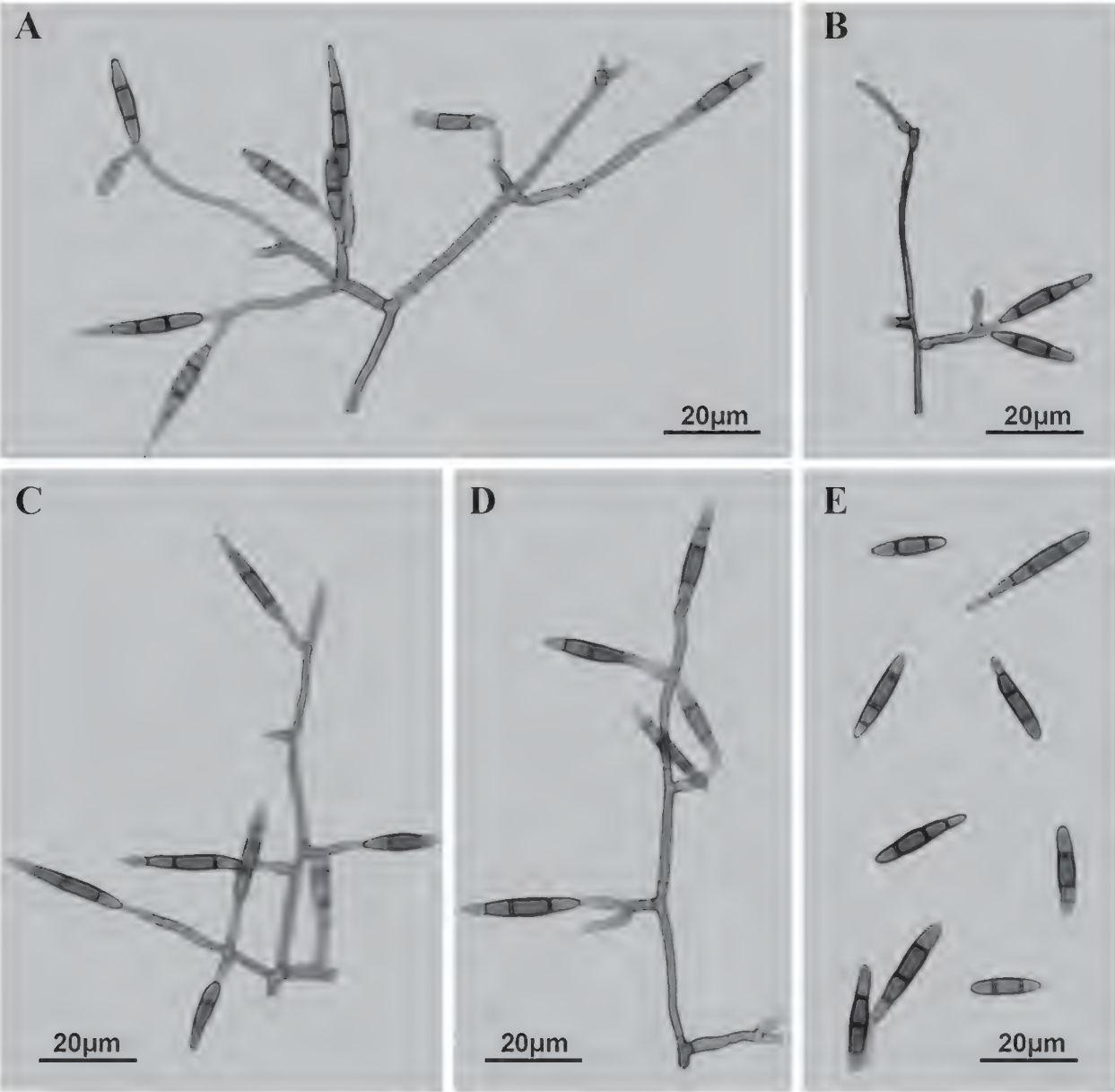


FIG. 1. *Dactylella yoaniae*. A–D. Conidiophores with conidia. E. Conidia.

Taxonomic description

*Dactylella yoaniae* Y.D. Zhang & X.G. Zhang, sp. nov.

FIGURE 1

MYCOBANK MB 518743

*Coloniae in substrato naturali effusae, pallide brunneae. Mycelium hyalinis, hyphae ramosae, pallide brunneae, septata, 3–4 µm crassis compositum. Conidiophora ex apice lateribusque hypharum oriunda, erecta, simplicia vel ramificata, incolorata, 1–4-septata, 11–77 µm longa, 3–4 µm lata ad basim, 2–3 µm lata ad apice. Conidia singula in apice conidiophori oriunda, fusiformis vel clavata, basi truncata, holoblastica, pallide brunneae, terminales, laevia, 2–4-septata, praecipue 3 septata, 18–34 × 3–5.5 µm.*

**HOLOTYPE:** on dead branches of *Yoania japonica* Maxim. (Orchidaceae), Longjingshan, Fujian Province, China. Aug. 11. 2009, Y.D. Zhang, HSAUP H3153 (isotype HMAS 144867).

**ETYMOLOGY:** in reference to the host genus, *Yoania*.

Colonies on the natural substratum, effuse, pale brown. Mycelium hyaline, hyphae flexuous and composed of branched, pale brown, septate, 3–4 µm thick. Conidiophores terminally and laterally on the hyphae, erect, simple or with several branches, colourless, 1–4-septate, 11–77 µm tall, 3–4 µm wide at the base, gradually tapering upward to 2–3 µm at the tip. Conidia formed singly at the apex of the conidiophores and on short branches, fusiform to clavate, truncate at the base, holoblastic, pale brown, smooth-walled, 2–4-septate, mainly 3-septate, 18–34 × 3–5.5 µm, median cells brown, the basal and apical cell becoming gradually paler.

The fungus is placed in the genus *Dactylella* based on its conidial shape and the multiseptate, single conidia. The conidia of *D. yoaniae* resemble those of *D. arnaudii* (Yadav 1960), *D. heptameres* (Drechsler 1943), and *D. clavata* (Gao et al. 1995) in having a similar conidial shape and conidiophore branches. However, the conidia of *D. yoaniae* are smaller than those of *D. arnaudii* (54–(69)–88 × 4.5–(7)–10 µm) and *D. heptameres* [(33–42)–55 × 7.5–(8.5)–9 µm]. In *D. heptameres* the conidia are 3–6-septate (mainly 6-septate) compared to the 2–4-septate (mainly 3-septate) conidia of *D. yoaniae*. *Dactylella clavata* has broader (4–(6)–8 µm) conidia, mainly 3–5-septate. In addition, in *D. yoaniae* the conidial basal and apical cells gradually become paler, which differs from other three species.

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## MYCOTAXON

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***Galerella nigeriensis* (Agaricales),  
a new species from tropical Africa**

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**Abstract** — A new species, *Galerella nigeriensis*, from southwestern Nigeria is described. It is characterized by a strongly plicate, dry, yellowish to orange brown pileus, whitish veil on pileus and stipe base, white and pubescent stipe, thick-walled, mostly flattened spores, tibiiform to lageniform cheilocystidia, and presence of hymenophysalides (recorded for the first time in the genus *Galerella*). Black and white photographs of basidiomata and microscopic elements accompany the description. *G. nigeriensis* is compared to related species and a worldwide diagnostic key to the genus *Galerella* is provided.

**Key words** — *Basidiomycota*, biodiversity, *Bolbitiaceae*, mycobiota, taxonomy

## Introduction

The third author conducted a field research of Nigerian mycobiota during the rainy season from June to August 2008. Among collected samples, we discovered a new species of *Galerella* that we describe here. *Galerella* Earle is a small genus of the family *Bolbitiaceae* Singer with five already known and well documented species: *G. fibrillosa* Hauskn., *G. floriformis* Hauskn., *G. microphues* (Berk. & Broome) Pegler, *G. plicatella* (Peck) Singer, and *G. plicatelloides* Sarwal & Locq. (see Sarwal & Locquin 1983, Hausknecht & Contu 2003). *Galerella conocephala* (Bull. : Fr.) Bon is considered a doubtful species by Hausknecht & Contu (2003) because of unclear interpretation and the lack of a holotype and recent material. *Galerella* species are saprotrophs, growing mostly on soil, but also on decaying twigs or wood. All species are rare (recorded only once or twice except *G. plicatella*). They are distributed throughout tropical and/or subtropical zone (including Mediterranean area), while *G. plicatella* also occurs



in areas with a continental climate. Morphologically, *Galerella* is characterized by a hymeniform pileipellis, rusty brown spore print, mainly dry and strikingly plicate-sulcate pileus (as in many *Coprinus* species), and by the absence of lecythiform cystidia (Horak 1968, Singer 1986, Hausknecht & Contu 2003). Although most authors consider *Galerella* an independent genus (Horak 1968, 2005, Moser 1983, Pegler 1986, Singer 1986, Bon 1992, Hausknecht & Contu 2003), some authors include *Galerella* in *Conocybe* Fayod s.l. (Watling 1982, as a subgenus) or *Pholiotina* Fayod (Arnolds 2005). In order to better understand the phylogenetic relationships between *Galerella* and related genera, molecular analyses are required.

### Materials and methods

The holotype description is based on one collection containing seven basidiomata, which were photographed in the field. Color codes in the macroscopic description (given in brackets) are cited according to Kornerup & Wanscher (1981). Microscopic features were observed using a light microscope (brightfield and phase contrast) with magnification up to 1500× and photographed with a digital camera. Description and photographs of microscopic characters were made from rehydrated dried specimens mounted in 2.5% potassium hydroxide (KOH) solution. Basidiospore color was also observed in H<sub>2</sub>O and 10% NH<sub>4</sub>OH. Basidiospore measurements were made from the mounts of lamellae and based on calibrated digital photographs: only mature spores (determined by color and appearance) were measured. The width of germ-pore was measured as inner distance between spore walls at the spore apex. A total number of 120 randomly selected basidiospores from two mature basidiomata were measured (60 in frontal view, 60 in side view). Spore measurements are given as: (min.) stat. min. – av. – stat. max. (max), where “min.” = minimum (lowest measured value), “stat. min.” = statistical minimum (arithmetic average minus two times standard deviation), “av.” = arithmetic average, “stat. max.” = statistical maximum (arithmetic average plus two times standard deviation), “max.” = maximum (highest measured value). Standard deviations (SD) of spore length, breadth, and width are also given. The length/breadth ratio of spores (frontal view) is given as the “Qf” value (min. – av. – max.) and length/width ratio of spores (side view) is given as the “Qs” value (min. – av. – max.). Holotype and accompanied data are deposited at the Croatian National Fungarium in Zagreb (CNF).

The term hymenophysalides is used according to Cléménçon (1997, 2004) for sterile, short, turgescient cells that surround the basidia (present in hymenium of some *Agaricales*), also called pseudoparaphyses, brachycystidia, brachybasidioles, or pavement cells. Comparison of *G. nigeriensis* with similar taxa and the diagnostic key of *Galerella* species are based on the descriptions



FIGS 1–2. Basidiomata of *Galerella nigeriensis* in situ. Bars = 5 mm.

and illustrations in the following literature: Horak 1968, Sarwal & Locquin 1983, Pegler 1986, Thomas et al. 2001, Horak & Hausknecht 2002, Arnolds

& Hausknecht 2003, Hausknecht & Contu 2003, Hausknecht et al. 2004, Hausknecht 2009.

## Taxonomy

*Galerella nigeriensis* Tkalčec, Mešić & Čerkez, sp. nov.

FIGS 1–10

MYCOBANK MB 518311

*Pileus* 14–17 mm *latus*, *valde plicatus*, *siccus*, *pallide flavido-brunneus vel aurantio-brunneus*. *Velum praesens*, *filamentosum*, *albicans*. *Lamellae anguste adnatae*, *ferrugineae*. *Stipes* 26–32 × 1–1.5 mm, *pubescens*, *albus*. *Sporae* (6.9–)7.3–8.8–10.4(–10.7) × (5.1–)5.3–6.1–6.8(–6.9) × (4.5–)4.6–5.3–6.0(–6.2) μm, *plerumque lentiformes*, *crasse tunicatae*, *in KOH ferrugineae*. *Hymenophysalides praesentes*, *cheilocystidia* (25–)30–65 × 8–14(–17) μm, *tibiiformia vel lageniformia*, *pileocystidia et caulocystidia praesentia*, *fibulae abundantes*.

ETYMOLOGY: The species is named after Nigeria, the country of origin.

HOLOTYPE: NIGERIA, ONDO STATE: 11 km NW of Akure, 7°19'28" N, 5°7'31" E, alt. 400 m, 25 Jul 2008, leg. M. Čerkez (CNF 1/5859).

PILEUS 14–17 mm broad, broadly ellipsoid to oblong at first, later obtusely conical with a small papilla, pale yellowish brown to light orange brown, with darker, orange brown (6C8) to dark reddish brown (8E8, 9E8, 9F8) center, not hygrophanous, surface dull, dry, strongly plicate-sulcate up to 3/4 of the radius. VEIL white or whitish, only in some places light brown, densely fibrillose and covering the whole basidioma at first, in maturity remains at the center of the pileus as small patches and usually at the base of the stipe as a small volva-like remnants. LAMELLAE narrowly adnate, rather crowded ( $L = \text{ca. } 32$ ,  $l = 0\text{--}3$ ), broad (up to 2 mm), very thin, white at first, later pale to rusty brown, with paler to concolorous, slightly flocculose edge. STIPE 26–32 × 1–1.5 mm, in the lower part gradually thickening to the base (up to 3 mm wide), white to pale cream, entirely densely pubescent, weakly striate lengthwise, dry, fistulose. CONTEXT very thin, whitish in stipe, brownish in pileus when moist and whitish on drying. SMELL and TASTE not recorded. SPORE PRINT rusty brown.

BASIDIOSPORES [ $120/2/1$ ] (6.9–)7.3–8.8–10.4(–10.7) × (5.1–)5.3–6.1–6.8(–6.9) × (4.5–)4.6–5.3–6.0(–6.2) μm,  $SD = 0.76 \times 0.37 \times 0.35$ ,  $Q_f = 1.29\text{--}1.44\text{--}1.71$ ,  $Q_s = 1.51\text{--}1.69\text{--}2.02$ , variable in size and shape, ellipsoid, slightly angular to subhexagonal, ovoid, limoniform or subamygdaliform in frontal view, ellipsoid, oblong or amygdaliform in side view, mostly flattened, thick-walled (0.6–0.9 μm), with central to slightly eccentric, ± truncate, 0.6–1.4 μm wide germ-pore, rusty brown in KOH and  $\text{NH}_4\text{OH}$ , yellow brown in  $\text{H}_2\text{O}$ , non-amyloid and non-dextrinoid. BASIDIA 18–23 × 8–11 μm, 4-spored, clavate, hyaline, thin-walled, surrounded by 3–5 hymenophysalides. BASIDIOLES narrowly clavate to clavate. HYMENOPHYSALIDES 16–40 × 11–22(–30) μm, subglobose, sphaeropedunculate, ellipsoid or broadly clavate, hyaline, well





FIGS 3–10. *Galerella nigeriensis*. 3. Spores. 4. Basidium (phase contrast). 5. Cheilocystidia (phase contrast). 6. Hymenophyses and basidia (phase contrast). 7. Pileipellis near margin of the pileus (phase contrast). 8. Pileipellis near center of the pileus (phase contrast). 9. Pileocystidium. 10. Caulocystidia. Bars: 3, 6–8 = 10  $\mu\text{m}$ ; 4 = 5  $\mu\text{m}$ ; 5, 9, 10 = 20  $\mu\text{m}$ .

developed in mature basidiomata. LAMELLAR EDGE almost sterile (basidia very rare). CHEILOCYSTIDIA (25–)30–65  $\times$  8–14(–17)  $\mu\text{m}$ , tibiiform ( $\pm$  50%) with subcapitate to capitate apex 5–11  $\mu\text{m}$  broad or lageniform with 3–5  $\mu\text{m}$

wide neck, less often conical, thin-walled to slightly thick-walled ( $\leq 0.5 \mu\text{m}$ ), hyaline. PLEUROCYSTIDIA absent. HYMENOPHORAL TRAMA made of much branched, mostly strongly and irregularly inflated hyphae, hyaline, thin-walled to thick-walled ( $\leq 0.8 \mu\text{m}$ ),  $1\text{--}20\text{--}32 \mu\text{m}$  wide. PILEIPELLIS a hymeniderm, at the center of the pileus physalo-palisadoderm, regularly formed only in young basidiomata, elements mainly broadly to narrowly clavate, less often ellipsoid, obovoid, subcylindrical or narrowly utriform,  $9\text{--}50\text{--}63 \times 3.5\text{--}12\text{--}18 \mu\text{m}$ , thin-walled, subhyaline. Yellowish brown intracellular pigment present in the subpellis and the upper part of pileal trama. PILEOCYSTIDIA scattered, lageniform with very long neck to filiform, hyaline, thin-walled,  $50\text{--}250 \times 6\text{--}17 \mu\text{m}$ , upper part  $3\text{--}5 \mu\text{m}$  broad. STIPITPELLIS a cutis, made of parallel, thin-walled, hyaline,  $2\text{--}10 \mu\text{m}$  wide hyphae. CAULOCYSTIDIA very variable in size and shape,  $10\text{--}330 \times 3\text{--}15 \mu\text{m}$ , mostly filiform or lageniform (often with very long neck), but also tibiiform, subcylindrical, clavate, ellipsoid or irregularly shaped, sometimes with horizontally elongated base, thin-walled, hyaline. VEIL made of elongated, occasionally branched, thin-walled, hyaline,  $1.5\text{--}4\text{--}6.5 \mu\text{m}$  wide hyphae. CLAMP CONNECTIONS abundant in all tissues.

HABITAT — Gregarious, lignicolous, on a very rotten stump at the edge of a heavily disturbed secondary tropical forest (with *Theobroma cacao*, *Musa* sp., *Elaeis guineensis*).

DISTRIBUTION — Known only from the type locality in Nigeria.

REMARKS — *Galerella nigeriensis* is characterized by a strongly plicate-sulcate, completely dry, pale yellowish brown to light orange brown pileus with a darker center, whitish veil on pileus and stipe base, white and pubescent stipe, thick-walled, mostly flattened and often somewhat angular basidiospores, tibiiform to lageniform cheilocystidia, and presence of hymenophysalides. Hitherto, hymenophysalides have been recorded only in the genera *Bolbitius* Fr., *Conocybe*, *Coprinus* Pers. s.l., and *Leucocoprinus* Pat. (Cléménçon 2004). Although our new species share this character with all *Bolbitius* and some *Conocybe* species, we placed our taxon in the genus *Galerella* on the basis of its strongly plicate-sulcate and completely dry pileus, well developed universal veil, and the absence of lecythiform cystidia. *Bolbitius* species have viscid pilei and lack universal veils, while *Conocybe* species have smooth or rugulose pilei, lecythiform cystidia, and lack universal veils. On the other hand, the presence of a delicate universal veil that covers the entire pileus in young stages was recorded by Hausknecht & Contu (2003) in three other *Galerella* species (*G. fibrillosa*, *G. floriformis*, and *G. plicatella*).

*Galerella nigeriensis* can be easily differentiated from other species in the genus by the presence of hymenophysalides and abundant tibiiform cheilocystidia (lacking in other *Galerella* species). *Pholiotina sulcata* Arnolds



& Hauskn. has until recently been mistaken for *G. plicatella* by European and probably Asian authors due to its pileus that varies from weakly striate to irregularly plicate-sulcate (Arnolds & Hausknecht 2003, Hausknecht 2009, Hausknecht et al. 2009). *Pholiotina sulcata* lacks hymenophysalides, tibiiform cheilocystidia, and a veil. The most important differences among world species of *Galerella* are presented in a diagnostic key.

Key to the world species of *Galerella*

- 1. Cheilocystidia absent ..... 2
- 1. Cheilocystidia present, well differentiated, and abundant ..... 3
- 2. Spores 11–16.5 × 7–10 µm, with germ-pore, thick-walled ..... *G. plicatelloides*
- 2. Spores 7–11 × 3.5–4 µm, without germ-pore, thin-walled ..... *G. floriformis*
- 3. Hymenophysalides present and well developed in mature basidiomata, cheilocystidia tibiiform and lageniform (in approximately equal proportion) ..... *G. nigeriensis*
- 3. Hymenophysalides absent, cheilocystidia not tibiiform (mostly lageniform, only sometimes with slightly broadened apex) ..... 4
- 4. Cheilocystidia ≤35 µm long, pileus whitish ..... *G. microphues*
- 4. Cheilocystidia ≤50(–65) µm long, pileus pale yellowish- to orange- or reddish-brown ..... 5
- 5. Spores thin- to slightly thick-walled, cheilocystidia 6–11(–16.5) µm broad ..... *G. plicatella*
- 5. Spores distinctly thick-walled, cheilocystidia 10–20 µm broad ..... *G. fibrillosa*

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We would like to thank Marco Contu for sending us the photograph of *G. plicatella* and to Vesna Lopina for her help with the Latin description. We are very grateful to Anton Hausknecht (Fakultätszentrum für Biodiversität der Universität Wien, Austria) and Dr. Vagner Gularte Cortez (Universidade Federal do Paraná, Brazil) for their critical reviews of the manuscript.

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## MYCOTAXON

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**Studies of *Exobasidium* new to China:  
*E. rhododendri-siderophylli* sp. nov. and *E. splendidum***

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**Abstract**—A new species, *Exobasidium rhododendri-siderophylli* causing leaf hypertrophy on *Rhododendron siderophyllum*, is described and a new Chinese record, *Exobasidium splendidum* on *Vaccinium fragile*, are reported from Yunnan Province, China. The new species is characterized by symptoms, number of sterigmata, and short germ tubes. Molecular sequence analyses of 22 *Exobasidium* species reveal that phylogenetic relationships within *Exobasidium* correspond to the host plants and symptoms.

**Key words**—*Exobasidiomycetes*, molecular analysis, taxonomy

A new species of *Exobasidium* on *Rhododendron siderophyllum* was collected from Yunnan Province. The host plant belongs to the subfamily *Rhododendroideae* of *Ericaceae*. The *Exobasidium* species is parasitic on young leaves and fruit, causing hypertrophy. The diseased leaf is almost wholly hypertrophied, pale yellow, and 2–3.3 cm long, 0.5–1.8 cm wide, and 2.5 mm thick; when mature, the underside is covered with a white hymenium. A transverse section of a diseased leaf shows a differentiation between the palisade and mesophyll cells, but it is not clear. The diseased fruit is entirely hypertrophied, 1.8 × 1.3 cm, and also covered with white hymenium when mature. The new species — characterized by the described symptoms, possession of 3–7 sterigmata, and short germ tubes — is described as:

***Exobasidium rhododendri-siderophylli*** ZhenYing Li & L. Guo, sp. nov.

MYCOBANK MB 518411

FIGS. 1–4

*Hymenium hypophyllum*. *Basidia* hyalina, cylindrica vel clavata, 5–9 µm lata, terminaliter 3–7 sterigmatibus 5–6(–7) × 1–1.5(–1.8) µm praedita. *Basidiosporae* ellipsoideae vel

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\*corresponding author

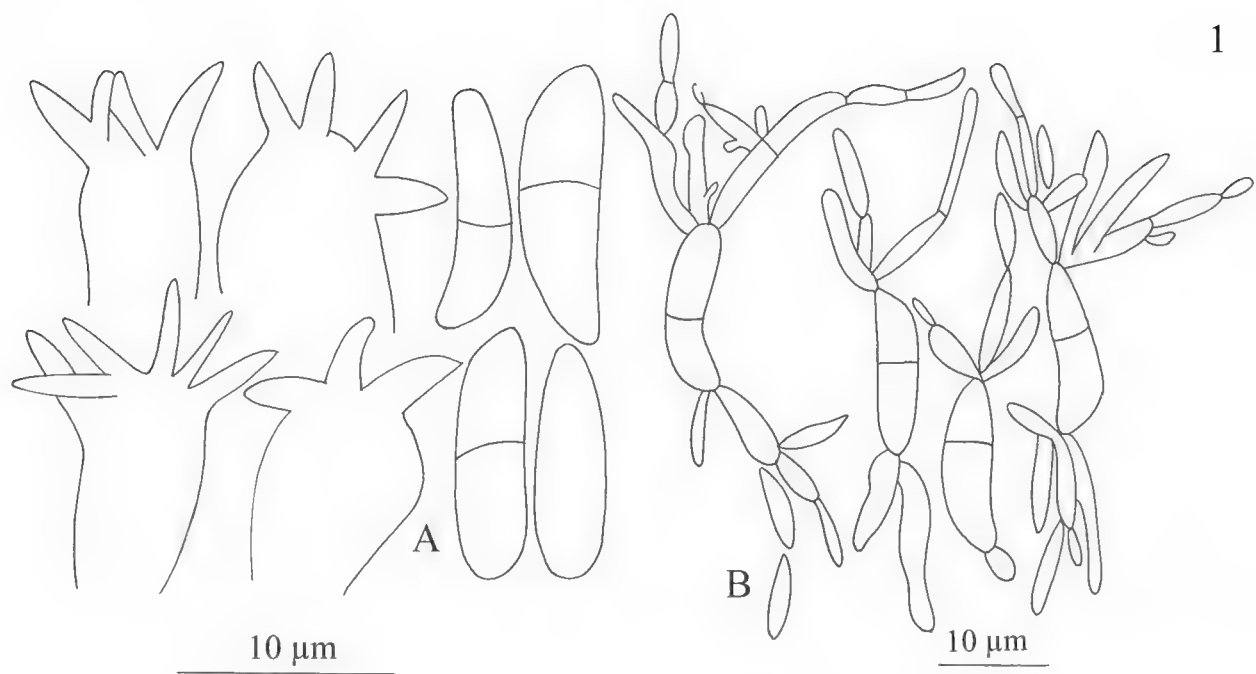


FIG. 1. Line drawings of *Exobasidium rhododendri-siderophylli* on *Rhododendron siderophyllum* (HMAS 183424, holotype). A. Basidia, sterigmata and basidiospores. B. Germinating basidiospores.

*clavatae, interdum curvae, (12–)13–15(–18.5) × 3–4 µm, hyalinae, leves, primo continuae, dein 1-septatae.*

TYPE: On *Rhododendron siderophyllum* Franch. (*Ericaceae*), Yunnan: Luquan, alt. 2520 m, 1.VII.2006, Z.Y. Li & L. Guo 339, HMAS 183424 (holotype).

Hymenium hypophyllous. Basidia hyaline, cylindrical or clavate, 5–9 µm diam., with 3–7 sterigmata. Sterigmata conical, 5–6(–7) × 1–1.5(–1.8) µm. Basidiospores ellipsoidal or clavate, occasionally curved, (12–)13–15(–18.5) × 3–4 µm, hyaline, smooth, at first continuous, then 1-septate.

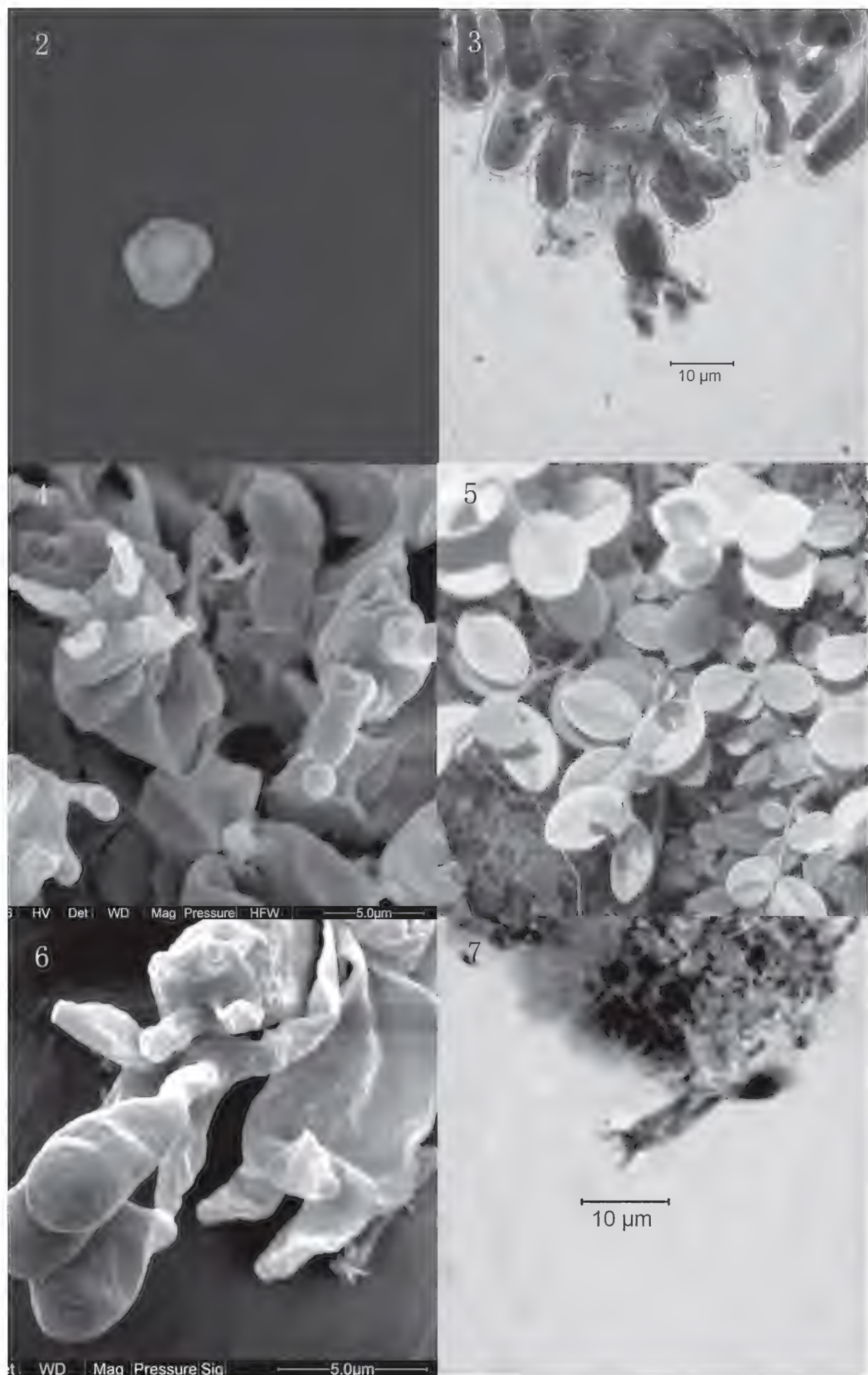
Colonies on potato dextrose agar (PDA) grew slowly, to a maximum 8 mm diameter after 21 days incubation at 25°C. The colony was pale yellow, composed of conidia. Conidia bacilliform, 5–7.5 × 1–2 µm.

ADDITIONAL SPECIMENS EXAMINED: On *Rhododendron siderophyllum* Franch. (*Ericaceae*), Yunnan: Luquan, alt. 2520 m, 1.VII.2006, Z.Y. Li & L. Guo 338, HMAS 183429 (paratype); Z.Y. Li & L. Guo 336, HMAS 183428 (paratype). On *Rhododendron tatsienense* Franch. (*Ericaceae*), Yunnan: Luquan, alt. 2530 m, 1.VII.2006, Z.Y. Li & L. Guo 329 HMAS 183437 (paratype).

REMARKS: Morphologically, *Exobasidium rhododendri* (Fuckel) C.E. Cramer (Nannfeldt 1981) on *Rhododendron ferrugineum* L. has similarly sized

FIGS. 2–4. *Exobasidium rhododendri-siderophylli* on *Rhododendron siderophyllum* (HMAS 183424, holotype). 2. Colony on PDA. 3. Basidium, sterigmata and basidiospores as seen by LM. 4. Basidia and sterigmata as seen by SEM. FIGS. 5–7. *Exobasidium splendidum* on *Vaccinium fragile* (HMAS 183436). 5. Symptoms. 6. Basidia, sterigmata and basidiospores as seen by SEM. 7. Basidium and sterigmata as seen by LM.





basidiospores ( $12\text{--}15 \times 2.5\text{--}4\ \mu\text{m}$ ) but differs from *E. rhododendri-siderophylli* in that it causes galls.

*Exobasidium splendidum*, discovered in Yunnan Province, is a new Chinese record. It is parasitic on *Vaccinium fragile*, causing leaf spots, usually 1(–2) on each leaf. The upper side of the diseased parts is slightly concave and red to pale red, and the underside becomes covered with white hymenium during maturation. The leaf spots can be 3.5–5.5 mm in diam. Transverse sections of the diseased leaf show clear differentiation of the palisade and mesophyll cells. There is no hypertrophy and hyperplasia of plant cells.

*Exobasidium splendidum* Nannf., Symb. Bot. Upsal. 23(2): 58, 1981.      FIGS. 5–8

SPECIMEN EXAMINED—On *Vaccinium fragile* Franch. (*Ericaceae*), Yunnan: Yangbi, Shangjie, Mopandi, alt. 2350 m, 14.IX.2005, Z.Y. Li, L. Guo & N. Liu 117, HMAS 183436.

Hymenium hypophyllous, white. Basidia hyaline, cylindrical, 4–8  $\mu\text{m}$ , with 2–4 sterigmata. Sterigmata conical,  $3\text{--}5 \times 1\text{--}2\ \mu\text{m}$ . Basidiospores cylindrical, clavate or obovoid, often curved,  $(7\text{--})9\text{--}14(\text{--}16) \times 3\text{--}4.2(\text{--}5)\ \mu\text{m}$ , hyaline, smooth, at first continuous, then 1–3-septate.

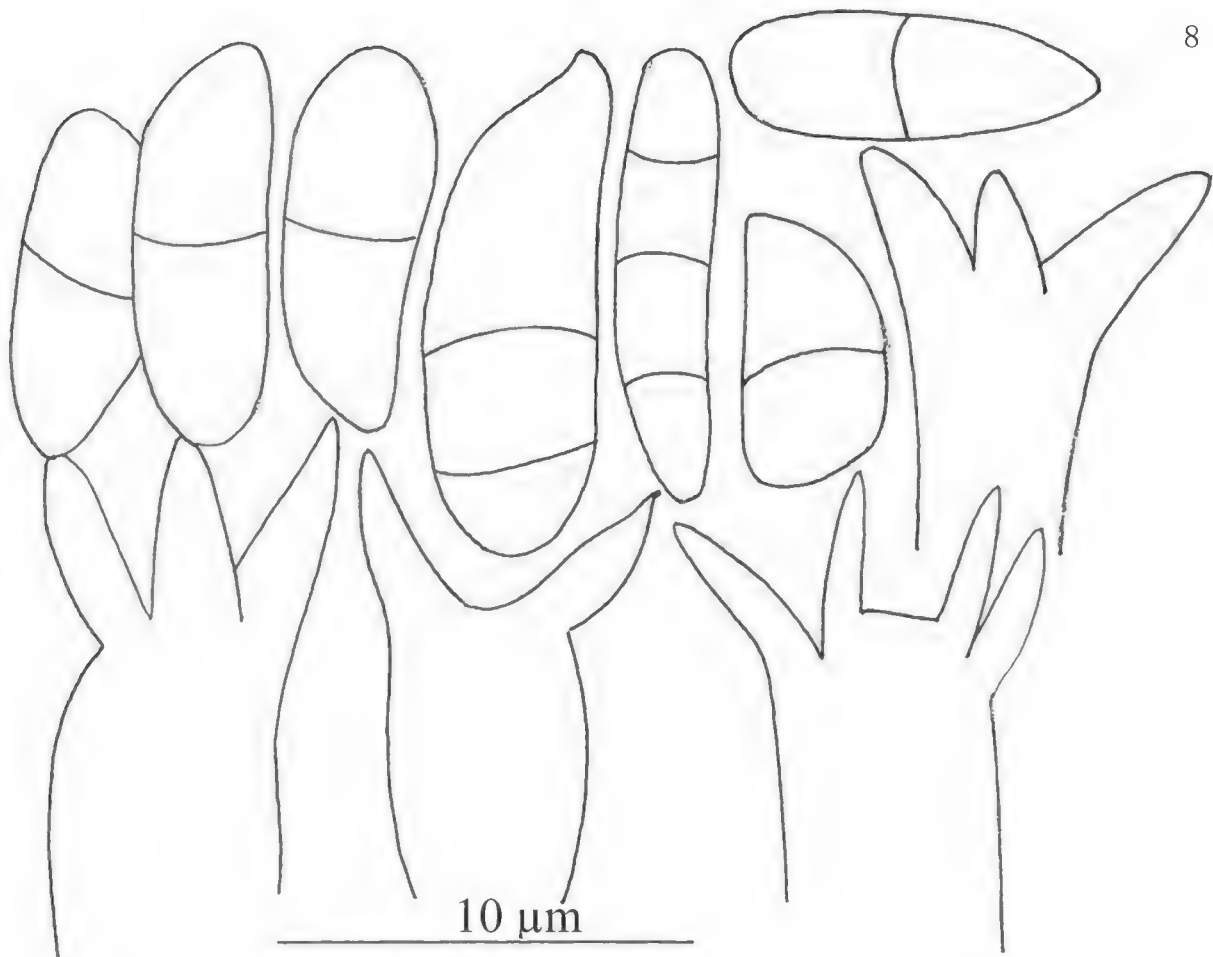


FIG. 8. Line drawings of *Exobasidium splendidum* on *Vaccinium fragile* (HMAS 183436).  
A. Basidia, sterigmata and basidiospores. B. Germinating basidiospores.

Thirty-three species of *Exobasidium* have been reported in China (Sawada 1922, Teng 1963, Tai 1979, Guo et al. 1991, Zang 1996, Li & Guo 2006a,b, 2008a,b, 2009a,b), including the two species recorded in this paper.

For phylogenetic analyses, the partial nrDNA-LSU (LSU) and ITS1-5.8S-ITS2 (ITS) genes were sequenced (White et al. 1990). Thirty-one sequences of 43 isolates (22 species) (TABLE 1), including 14 sequences (11 species) downloaded from Genbank, were used. Seventeen isolates (11 species) were collected by the authors. All strains collected by the authors were deposited in China General Microbiological Collection Center (CGMCC) (TABLE 1), and all sequences generated in this study were submitted to GenBank. Two *Entyloma* species were used as outgroup.

TABLE 1. Materials used in analysis of the nrDNA-LSU and nrDNA-ITS rDNA sequences

TAXON	SYMPTOM	HOST	COLLECTION	GENBANK NO.	
				LSU	ITS
<i>E. bisporum</i>	leaf spots	<i>Eu. grayana</i> var. <i>glabra</i>	IFO9942	AB177598	AB180364
<i>E. camelliae</i>	fruit & leaf hypertrophy	<i>C. japonica</i>	MAFF238578	AB176712	AB180317
<i>E. canadense</i> *	leaf spots	<i>R. mariesii</i>	HMAS 173409	EU692791	EU692771
<i>E. cylindrosporum</i>	leaf spots	<i>R. sp.</i>	MAFF238608	AB178245	
<i>E. cylindrosporum</i>	leaf spots	<i>R. pulchrum</i>	MAFF238579		AB180318
<i>E. cylindrosporum</i> *	leaf spots	<i>R. sp.</i>	HMAS 183415	EU692795	EU692776
<i>E. euryae</i> *	galls	<i>C. oleifera</i>	HMAS 97947	EU692779	EU692759
<i>E. formosanum</i> *	galls	<i>R. delavayi</i>	HMAS 183418	EU692781	EU692775
<i>E. formosanum</i> *	galls	<i>R. sp.</i>	HMAS 183445	EU692796	EU692777
<i>E. gracile</i> *	leaf hypertrophy	<i>C. oleifera</i>	HMAS 140210	EU692780	
<i>E. gracile</i> *	leaf hypertrophy	<i>C. oleifera</i>	HMAS 140502		EU692761
<i>E. gracile</i>	leaf hypertrophy	<i>C. sasanqua</i>	TUK-E30	AB177592	
<i>E. gracile</i>	leaf hypertrophy	<i>C. sasanqua</i>	MAFF238586		AB180322
<i>E. inconspicuum</i>	leaf spots	<i>V. hirtum</i> var. <i>pubescens</i>	MAFF238616	AB177556	
<i>E. inconspicuum</i>	leaf spots	<i>V. hirtum</i> var. <i>pubescens</i>	MAFF238619		AB180350
<i>E. japonicum</i> *	leaf deform & hypertrophy	<i>R. pulchrum</i>	HMAS 172284	EU692788	EU692773
<i>E. japonicum</i> *	leaf hypertrophy	<i>R. simsii</i>	HMAS 175467	EU692790	EU692766
<i>E. japonicum</i> *	leaf deform & hypertrophy	<i>R. sp.</i>	HMAS 175457	EU692792	EU692772
<i>E. japonicum</i> *	leaf deform & hypertrophy	<i>R. sp.</i>	HMAS 175455	EU692793	EU692768

TABLE 1, concluded.

TAXON	SYMPTOM	HOST	COLLECTION	GENBANK NO.	
				LSU	ITS
<i>E. japonicum</i> *	leaf deform & hypertrophy	<i>R. sp.</i>	HMAS 175454	EU692794	EU692769
<i>E. japonicum</i>	leaf deform & hypertrophy	<i>R. obtusum</i> var. <i>kaempferi</i>	MAFF238826	AB178253	
<i>E. japonicum</i>	leaf deform & hypertrophy	<i>R. lateritium</i>	IFO30756		AB180370
<i>E. kunmingense</i> *	leaf spots	<i>L. ovalifolia</i>	HMAS 173147	EU692784	EU692763
<i>E. lushanense</i> *	leaf spots	<i>R. simsii</i>	HMAS 173148	EU692789	EU692767
<i>E. miyabei</i>	leaf spots	<i>R. dauricum</i>	MAFF238583	AB177550	
<i>E. miyabei</i>	leaf spots	<i>R. dauricum</i>	MAFF238595		AB180330
<i>E. nobeyamense</i>	witches' broom	<i>R. wadanum</i>	MAFF238583	AB180378	
<i>E. nobeyamense</i>	witches' broom	<i>R. wadanum</i>	MAFF238598		AB180332
<i>E. otanianum</i>	leaf spots		IFO9960	AB177600	
<i>E. otanianum</i>	leaf spots	<i>R. hyugaense</i>	MAFF238612		AB180344
<i>E. pentasporium</i>	witches' broom & leaf spots	<i>R. obtusum</i> var. <i>kaempferi</i>	MAFF238601	AB177567	
<i>E. pentasporium</i>	witches' broom & leaf spots	<i>R. obtusum</i> var. <i>kaempferi</i>	MAFF238179		AB180316
<i>E. pieridis-ovalifoliae</i>	leaf spots	<i>L. neziki</i>	IFO9961	AB177601	AB180367
<i>E. rhododendri</i>	galls	<i>R. ferrugineum</i>	R.B.2050	AF009856	
<i>E. rhododendri</i>	galls	<i>R. sp.</i>	CBS101457		DQ667153
<i>E. rhododendri-russati</i> *	galls	<i>R. russatum</i>	HMAS 183433	EU692797	EU692778
<i>E. rhododendri-siderophylli</i> *	leaf hypertrophy	<i>R. tatsienense</i>	HMAS 183437	EU692782	EU692762
<i>E. rhododendri-siderophylli</i> *	leaf hypertrophy	<i>R. siderophyllum</i>	HMAS 183428	EU692786	EU692765
<i>E. rhododendri-siderophylli</i> *	leaf hypertrophy	<i>R. siderophyllum</i>	HMAS 183429	EU692786	EU692764
<i>E. woronichinii</i>	leaf spots	<i>R. brachycarpum</i>	MAFF238825	AB178252	
<i>E. woronichinii</i>	leaf spots	<i>R. brachycarpum</i>	MAFF238617		AB180348
<i>E. yoshinagae</i>	leaf spots	<i>R. wadanum</i>	MAFF238606	AB177551	
<i>E. yoshinagae</i>	leaf spots	<i>R. reticulatum</i>	IFO9959		AB180365
<i>Entyloma ficariae</i>		<i>Ra. ficaria</i>		AY081013	
<i>Entyloma ficariae</i>		<i>Ra. ficaria</i>			AY081035
<i>Entyloma linariae</i>		<i>Linaria vulgaris</i>		AY860054	
<i>Entyloma linariae</i>		<i>Linaria vulgaris</i>			AY081041

\* = collected and sequenced by the authors.  
C.= *Camellia*, E.= *Exobasidium*, Eu.= *Eubotryoides*, L.= *Lyonia*, R.= *Rhododendron*, Ra.= *Ranunculus*,  
V.= *Vaccinium*.

Two sequence sets, both independently and combined, were analyzed following the Minimum Evolution method (ME) (Rzhetsky & Nei 1992). As

all the ME trees derived from the independent and combined ITS and LSU sequence analyses share similar topologies structure and main clades, only the ME tree based on the combined ITS and LSU analysis is shown (FIG. 9).

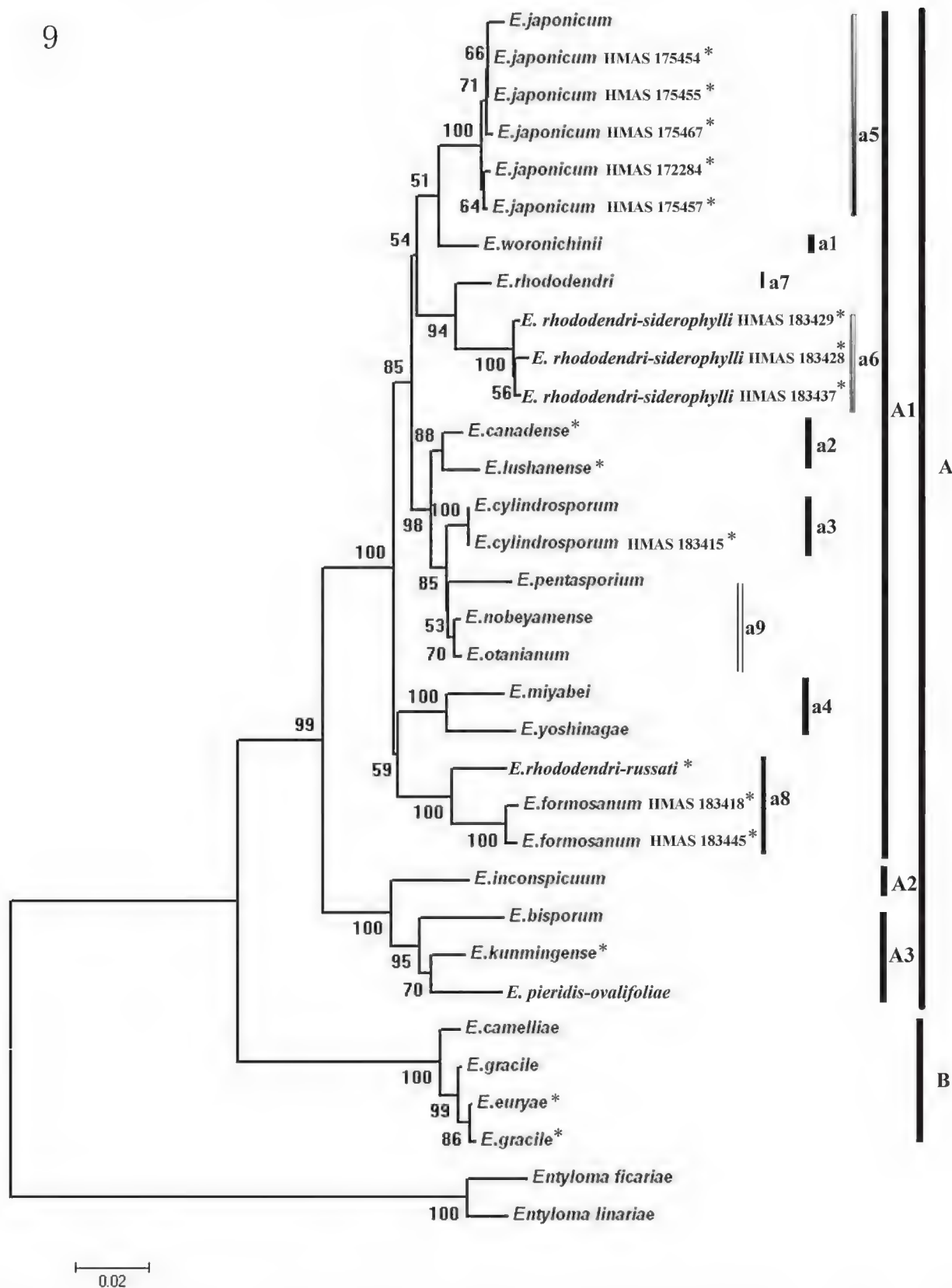


FIG. 9. ME tree based on analysis of nrDNA-ITS/nrDNA-LSU sequences. The numbers on the branches indicate bootstrap values, following the 50% majority rule. \* = collected and sequenced by the authors; *E.* = *Exobasidium*. Bar types correspond to the different symptoms, i.e. leaf spots (a1–a4), leaf hypertrophy (a5–a6), galls (a7–a8), and witches' broom (a9).



The combined tree is the most parsimonious following the 50% bootstrap majority-rule.

Two major clades (A–B) are identified in the ME tree: clade A consists of only the species parasitic on *Ericaceae*, while clade B contains species on *Theaceae*. Clade A includes three subclades: A1 on *Rhododendroideae* (*Rhododendron*), A2 on *Vaccinioideae*, and A3 on *Andromedoideae*. A1 encompasses nine small clades, including species causing different symptoms — a1–a4 causing leaf spots, a5–a6 leaf hypertrophy, a7–a8 galls, and a9 witches' broom.

Results of the molecular analyses indicate that the phylogenetic relationships within *Exobasidium* correspond to the host plants and symptoms. Host associations and symptoms should be regarded as important characteristics for morphological identification.

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**Biogeographical patterns in  
pyrenomycetous fungi and their taxonomy.  
1. The Grayan disjunction**

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**Abstract** — In this paper the biogeographical pattern known as the Grayan disjunction is discussed with respect to pyrenomycetous fungi. The importance of considering biogeographical data in taxonomy is emphasized. *Apiognomonina duschekiae* is described as a new species, *Biscogniauxia alnophila* is proposed as a new name for *B. mediterranea* var. *microspora*, and *Nemania sphaerostoma* is proposed as a new combination.

**Key words** — Ascomycota, biogeography, distribution

## Introduction

The importance of pyrenomycetous fungi in ecosystems as decomposer organisms cannot be overestimated, but many issues relating to their taxonomy, ecological preferences, and geographical patterns remain unclear on a global scale.

Many pyrenomycetous fungi are restricted to particular hosts, and this association suggests that each species follows the distribution of its substrates, at least within uniform climatic zones, such as the cold temperate, warm temperate, or tropical zones. Indeed, there are circumpolar, circum-boreal, and pan-tropical pyrenomycete species, which some might consider to represent the primary distribution patterns for these fungi. More limited and peculiar patterns have been not discussed or even suspected. As a result, although the Asian mycobiotas are not similar to the European mycobiota, mycologists often have applied European names to morphologically similar Asian fungi because they assume that fungi are widely distributed.

This paper discusses one biogeographical pattern that is usually termed the Grayan (Petersen & Hughes 2007) or Graysian (Tulloss 2005) disjunction in mycological literature. A number of plants and animals restricted to eastern North America where remnants of the ancient Tertiary flora persist can also occur in similar fragments of that flora in eastern Asia. Such a distribution, known as the famous “Asa Gray disjunction,” has been reported for species of fungi, primarily macrofungi (Hongo & Yokoyama 1978, Zang 1986, Wu & Mueller 1997, Yang 2000, Mueller et al. 2001) or lichenized fungi (Culberson 1972, Dey 1976, Wei & Biazrov 1991). An examination of the biogeographical patterns of pyrenomycetous fungi, which have not been considered previously, reveals a number of examples of Grayan distribution.

### Materials and methods

The specimens mentioned in this study were collected by the senior author over many years throughout eastern Russia and the eastern United States. The basic map was taken from the web site <http://commons.wikimedia.org> and modified with our data. Photographs of ascomata were obtained using a Nikon D40x digital camera.

### Non-vicariance pattern

Among the pyrenomycetous fungi are two species groups that demonstrate a Grayan distribution—those that occur in eastern Asia and eastern North America and those that display a vicariance pattern. Examples of the first group are *Fracchiaea callista* (Berk. & M.A. Curtis) Sacc. (FIG. 1), “*Diatrypella informis*” Ellis & Everh. (FIG. 2), *Graphostroma platystoma* (Schwein.) Piroz. (FIG. 3), and possibly *Nitschkia floridana* Fitzp. (Vasilyeva et al. 2010). *Graphostroma platystoma* occurs on dead branches of many kinds of trees, suggesting a wide distribution, but the fungus displays an affinity for eastern Asia and eastern North America. Similarly *Diatrype albopruinosa* (Schwein.) Cooke is found only in these two widely separated areas (FIG. 4); it has a broad tree host range in eastern North America (Rappaz 1987) but occurs only on *Padus avium* Mill. in eastern Russia. This is not the only example of an apparent substrate preference displayed by pyrenomycetous or loculoascomycetous fungi in eastern Russia.

As another example, *Byssosphaeria rhodomphala* (Berk.) Cooke, occurs in eastern Russia only on *Maackia amurensis* Rupr., *Phellodendron amurense* Rupr., and *P. sachalinense* (F. Schmidt) Sarg., whereas in North America this species is known mostly on *Populus* spp. and *Robinia pseudoacacia* L. (Barr 1990). Both *Populus* and *Robinia* are present in eastern Russia, yet they apparently never serve as hosts to *Byssosphaeria rhodomphala*. *Maackia* and *Robinia* are both members of the *Fabaceae*, unlike the more distantly related *Phellodendron* (*Rutaceae*) and *Populus* (*Salicaceae*). The preference of the same



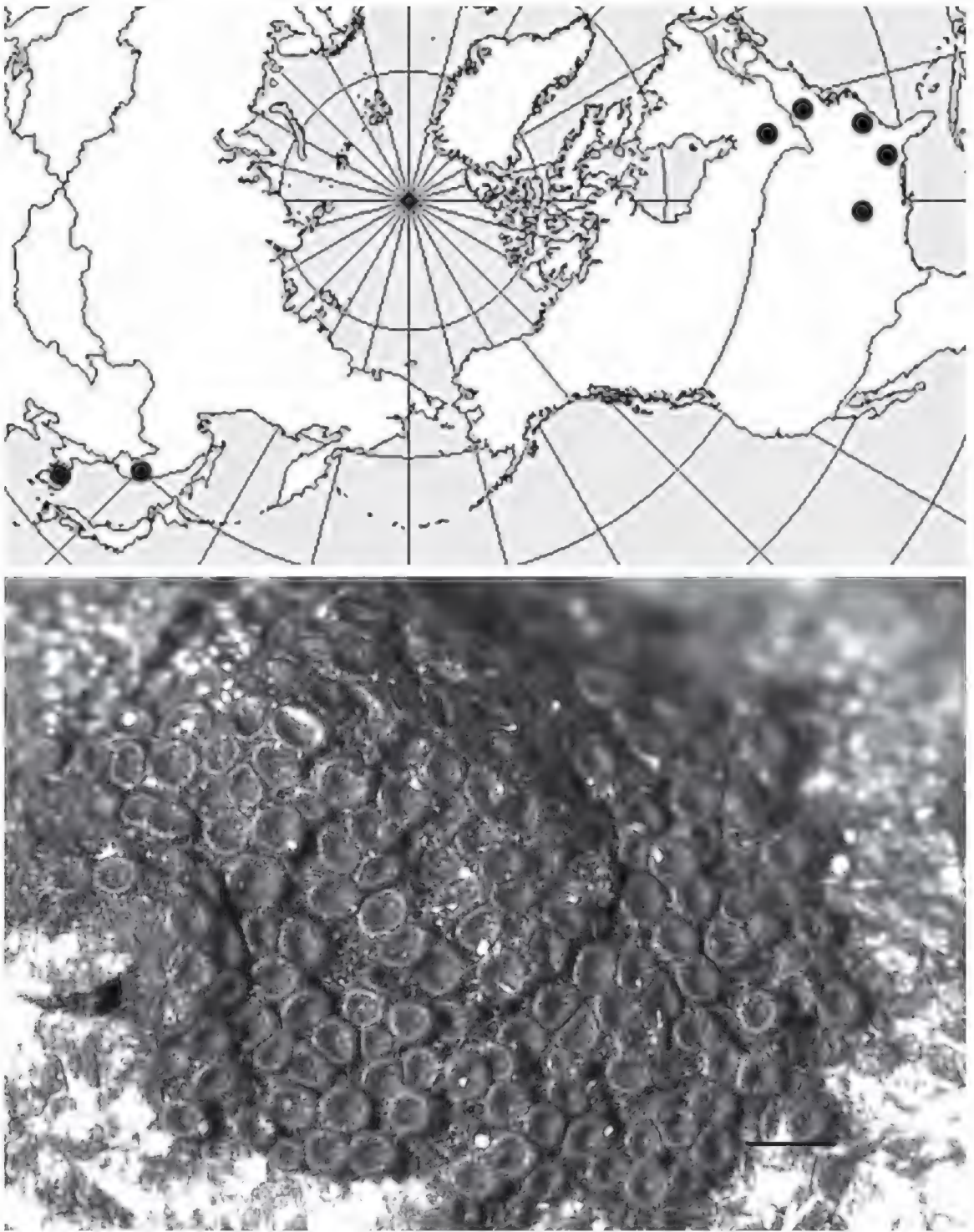


FIG. 1. Approximate biogeographical distribution of *Fracchiaea callista*. For North America, Nannfeldt (1975) cited the Alabama, Ontario, Pennsylvania, and South Carolina localities while the westernmost record thus far from Arkansas is supported by Vasilyeva's collection in the Buffalo National River; Connecticut, Maryland and Virginia are omitted. Localities in eastern Russia and South Korea are also based on the first author's own collections. Scale bar = 0.75 mm.

species for different hosts in different regions remains inexplicable — unless they are not the same species. If further studies prove them to be different species, they would represent a vicariance pattern in the Grayan distribution, discussed below.

A good example of Grayan disjunction is *Hypoxylon sphaeriosomum*, known earlier from the USA (Georgia, Ohio, and Pennsylvania; Miller 1961)



FIG. 2. Approximate biogeographical distribution of “*Diatrypella informis*”. Only two localities are indicated for eastern Russia, although this species is rather common on dead branches of *Carpinus cordata* Blume and is also found throughout the Primorsky Territory, including the Sikhote-Ainsky Nature Reserve, Kedrovaya Pad Biosphere Reserve, Ussuriysky Nature Reserve, the Vladivostok vicinity, and near Anisimovka (District Shkotovo). The eastern North American locality is based on Ellis & Everhart’s North American Fungi No. 2530 (“*Diatrypella informis* E. & E. n. sp. (TYPE), on dead *Carpinus*, London, Canada, Apr. 1890, J. Dearness”). Scale bar = 1 mm.

and recorded later from eastern Russia (FIG. 5). This species, which Ju & Rogers (1996) excluded from *Hypoxylon* (considering it to belong to *Euepixylon*), is treated herein as *Nemania sphaeriostoma*.

Some species that display an apparent Grayan distribution have been reduced to synonyms, although they are morphologically distinctive and have a restricted distribution. For instance, Ju et al. (1998) regarded *Biscogniauxia pezizoides* (Ellis & Everh.) Kuntze as synonymous with *B. repanda* (Fr.) Kuntze.



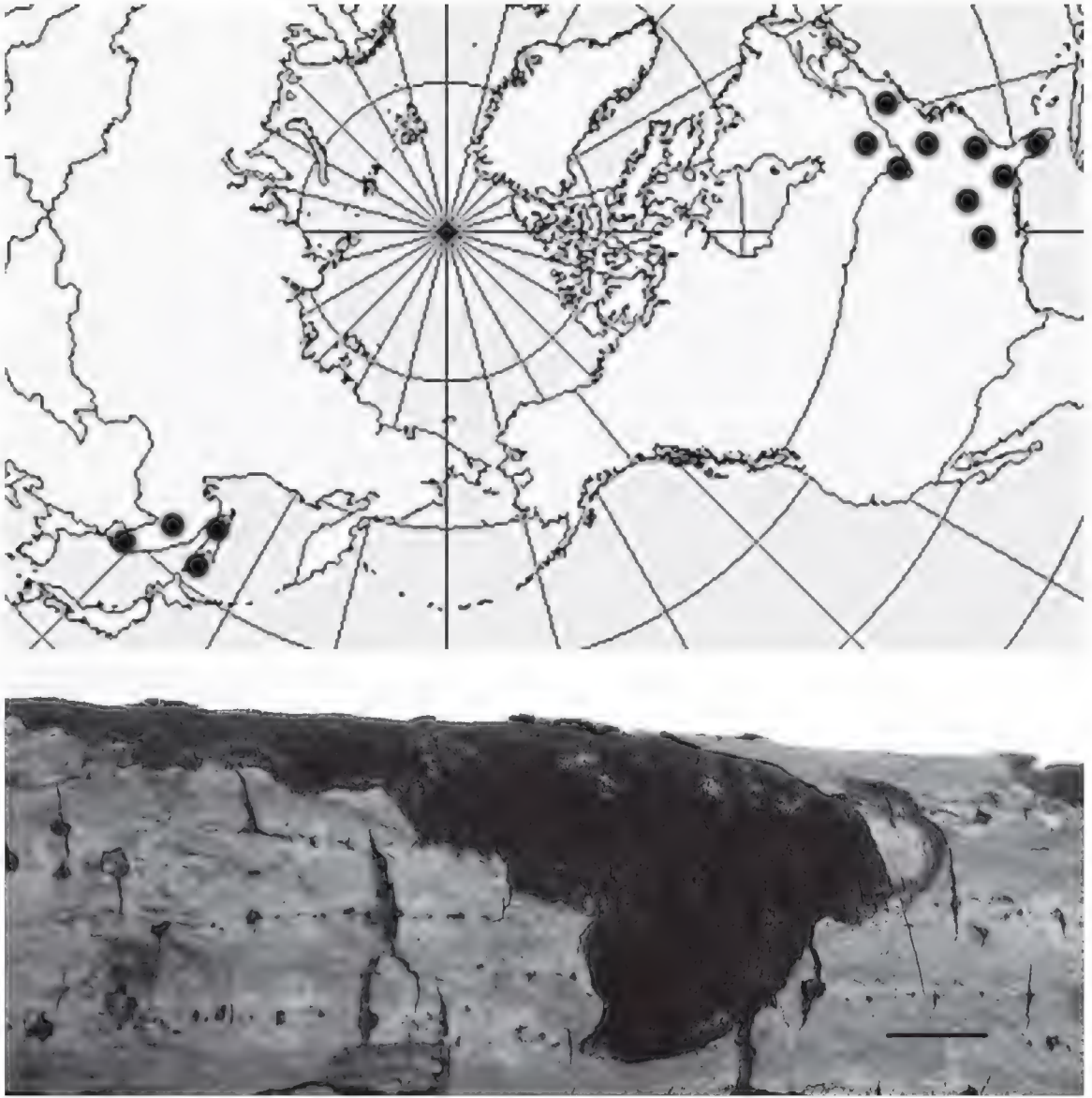


FIG. 3. Approximate biogeographical distribution of *Graphostroma platystoma*. North American localities listed by Pirozynski (1974) include Ontario and Quebec in Canada and Alabama, Arkansas, Florida, Massachusetts, Missouri, New Jersey, New York, North Carolina, Ohio, Pennsylvania, South Carolina, and Vermont in the USA. The eastern Russian specimens were collected in the Primorsky and Khabarovsk territories, the Amur Region, and on Sakhalin and Kunashir islands. Scale bar = 0.7 mm.

However these two names might just as easily represent different species that are restricted to different host plants (mostly *Ulmus* and *Sorbus*, respectively) and their occurrence on different continents has already been noted (Pouzar 1979). The later discovery of *B. pezizoides* in eastern Asia (Vasilyeva 1998) fits its distribution in the Grayan disjunction (FIG. 6). Another example is *Diaporthella platasca* (Peck) Wehm. (FIG. 7), first described from the Adirondack Mountains in eastern United States (Peck 1873) and later been shown (Wehmeyer 1933) to have smaller stromata and larger ascospores (16–23  $\mu\text{m}$  long) than the European species *D. aristata* (Fr.) Petr. (ascospores 13–16  $\mu\text{m}$  long). However, the two species were later confused and referred to *D. aristata* (Barr 1978, Chlebicki 2002). When *D. aristata* and *D. platasca* were found in eastern

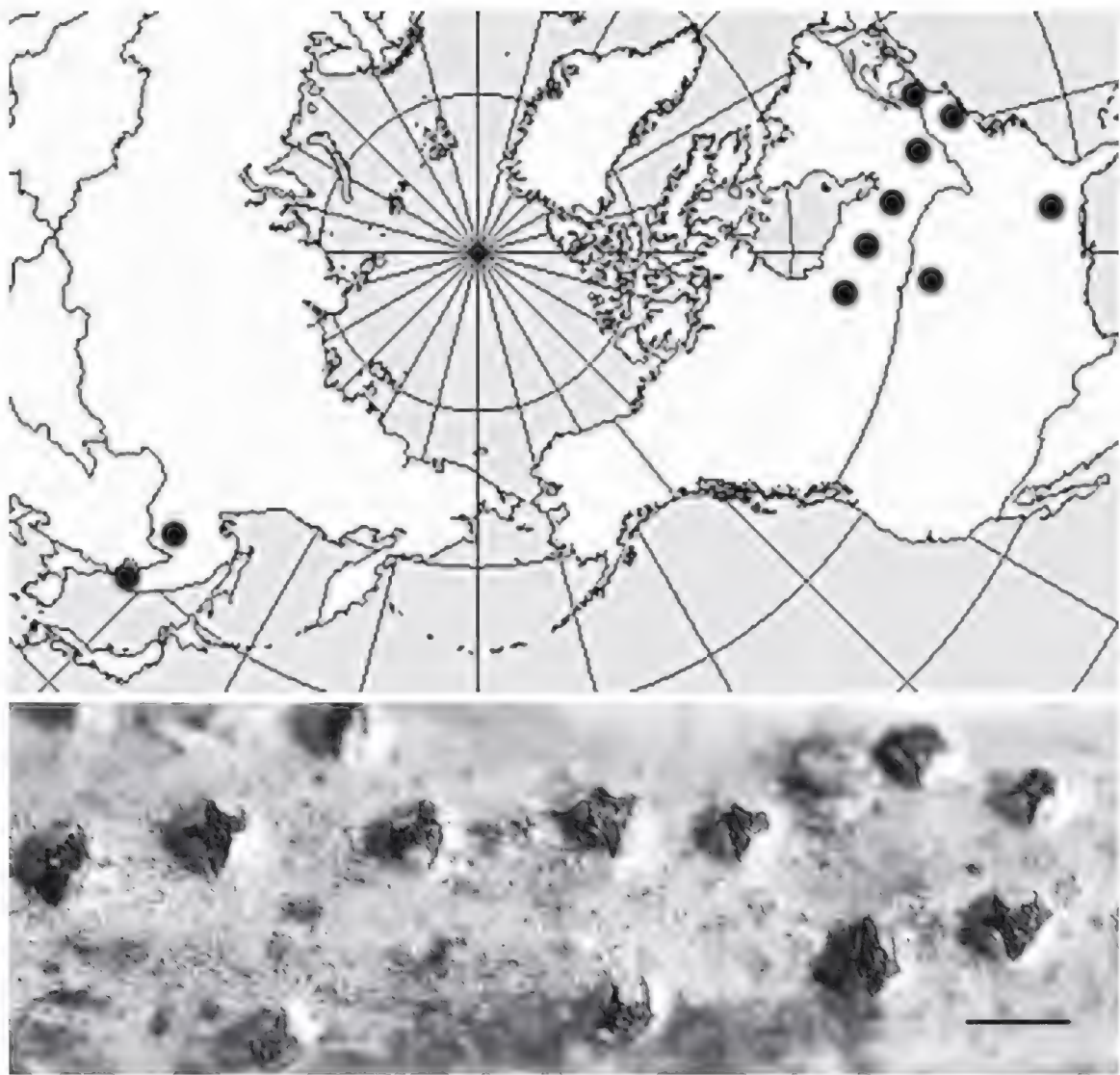


FIG. 4. Approximate biogeographical distribution of *Diatrype albopruinosa*. Some USA localities (Connecticut, the Dakotas, New Jersey, Mississippi) are from Rappaz (1987), and those for Canada (Manitoba, Ontario, Quebec, Saskatchewan) are taken from MycoBank ([www.mycobank.com](http://www.mycobank.com)). The distribution range of this species in North America extends more to the west than for many other species with a Grayan disjunction. Only two collections are known from eastern Russia (in the Primorsky Territory and the Amur Region), but the senior author also found *D. albopruinosa* in China (Heilongjiang Province). Scale bar = 1 mm.

Asia—on the Kamchatka Peninsula and Sakhalin Island, respectively—their differences became evident, not only with respect to morphology but also in their ecological preferences. *Diaporthella aristata* parasitizes living branches of birch trees (*Betula ermanii* Cham.), whereas *D. platasca* occurs on dead branches of low shrubs (*Betula middendorffii* Trautv. & C.A. Mey.).

While describing the genus *Diaporthella*, Petrak (1924) noted the parasitic nature of *D. aristata*. However, the particular kind of substrate (trees or shrubs, in this case) might be of no importance, since Chlebicki (2002) indicated that *D. aristata* (with typical ascospores 14–16 µm long) occurs on living and dead twigs of a very low shrub (*Betula nana* L.). When discussing the material of *D. aristata* examined from North America, Barr (1978) made reference only to



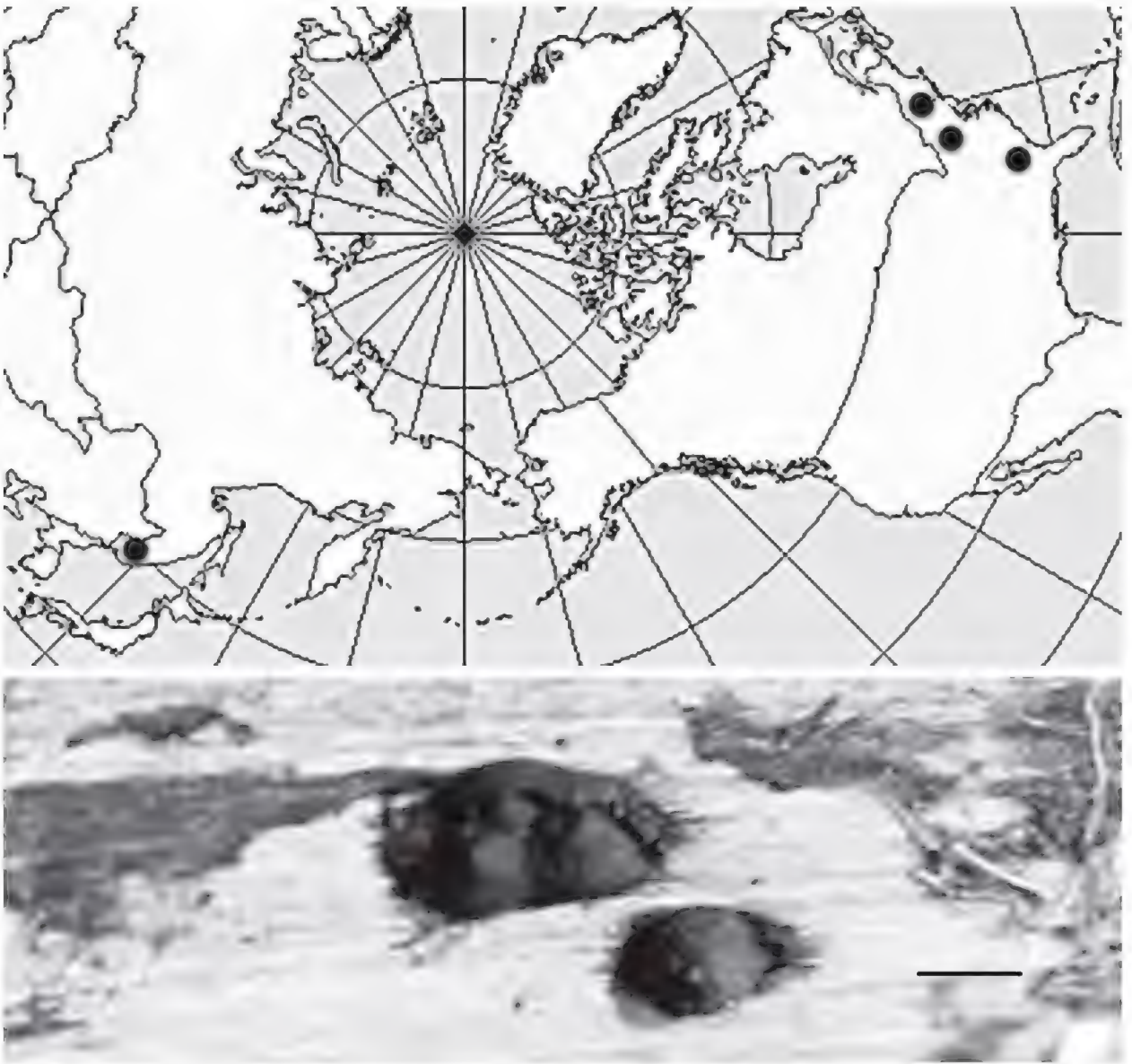


FIG. 5. Approximate biogeographical distribution of *Nemanium sphaeriosoma*. The USA localities are cited by Miller (1961). The circle on the map encompasses the two collections from the Ussuriysky and Lazovsky Nature Reserves (Primorsky Territory) in eastern Russia. Scale bar = 1.5 mm

the type of *D. platasca*; therefore, the occurrence of the true *D. aristata* in North America is unknown.

The focus upon the biogeographical pattern discussed here has forced a reconsideration of species concepts. For example, specimens of *Apiognomonia alniella* (P. Karst.) Höhn. on the dead leaves of *Alnus fruticosa* Rupr. from the Magadan region (Vasilyeva 1987) fit Barr's description of a species indicated as occurring on overwintered leaves of *Alnus* spp. in Europe and North America (Barr 1978). However, Barr's North American specimens were collected in Quebec and Maine, regions in the eastern portion of the continent that share so many species in common with eastern Russia.

Further investigations showed that most of the European specimens of *Apiognomonia alniella* in exsiccatae contain living leaves of *Alnus incana* (L.) Moench covered by extensive necrotic spots caused by a parasitic fungus,



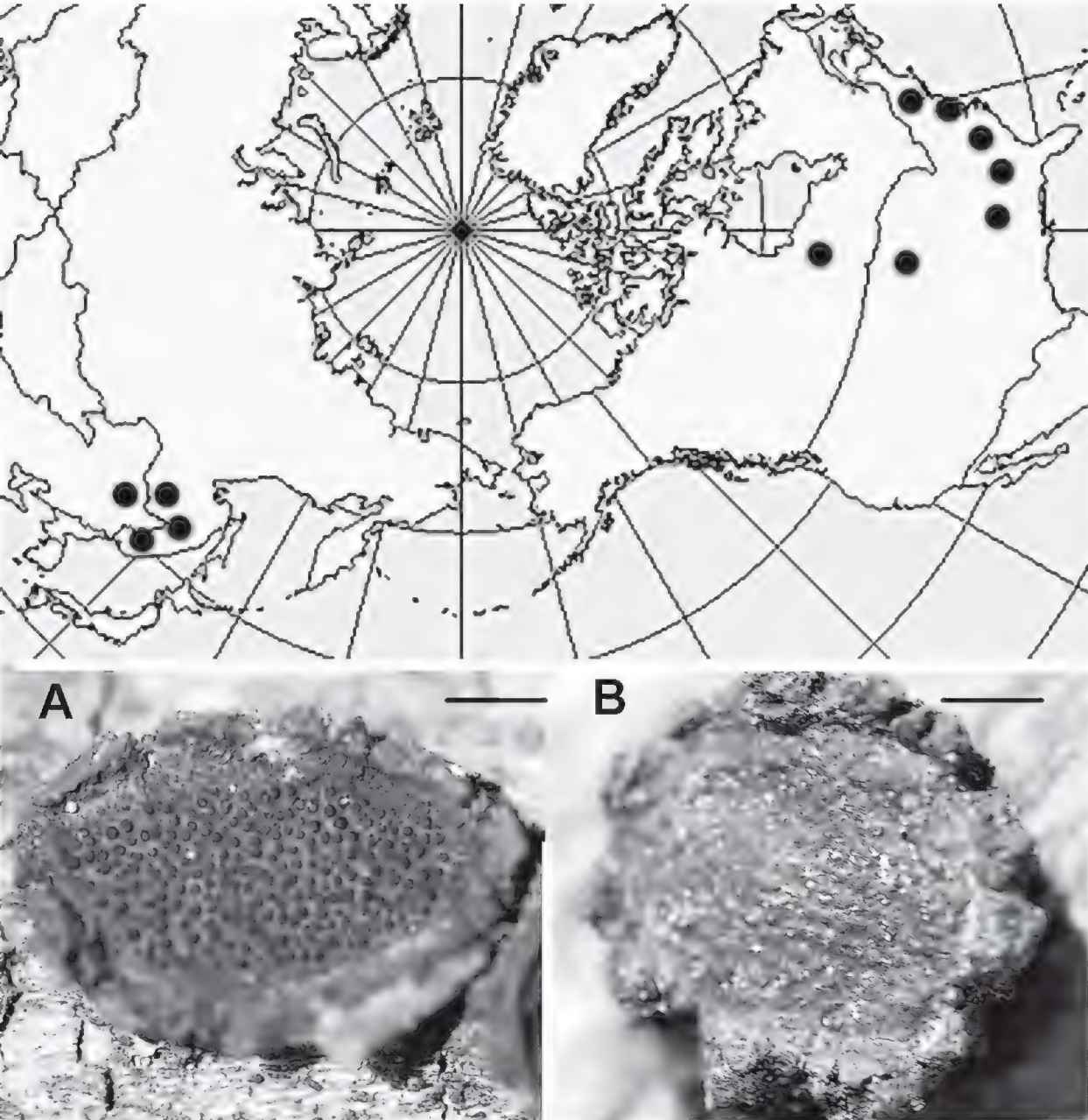


FIG. 6. Approximate biogeographical distribution of *Biscogniauxia pezizoides*. A - Stroma of *B. pezizoides* studded with the characteristic ostioles. B - Stroma of *B. repanda* [from western Russia, Leningrad Region, on *Sorbus aucuparia* L., D. Shabunin, VLA P-1429]. In North America, most localities (Delaware, Manitoba, Maryland, Nebraska, Virginia) are based on *B. pezizoides* specimens on *Ulmus* spp. (BPI collections: 595296, 595313, 595312, 595310, 595309); BPI 595300 from New York was collected on *Acer* sp., and the senior author has collected from *Acer* ap. in Tennessee (Great Smoky Mountains National Park) and from *Ulmus* sp. in Arkansas (Buffalo National River) as well as from *Acer mono* Maxim. and *Ulmus* spp. in eastern Russia and northeastern China (Heilongjiang province). Scale bars: 6A = 1.4 mm, 6B = 1.7 mm.

and the perithecia present are usually immature. We have observed exactly the same kind of a necrosis on living leaves of *A. hirsuta* (Spach) Turcz. ex Rupr. collected on the Kamchatka Peninsula. The immature perithecia were quite different from those on dead leaves of *A. fruticosa* in the Magadan region (FIG. 8). The immature state of the Kamchatka specimen did not allow us to make the proper comparison for a long time, but all the data available in the

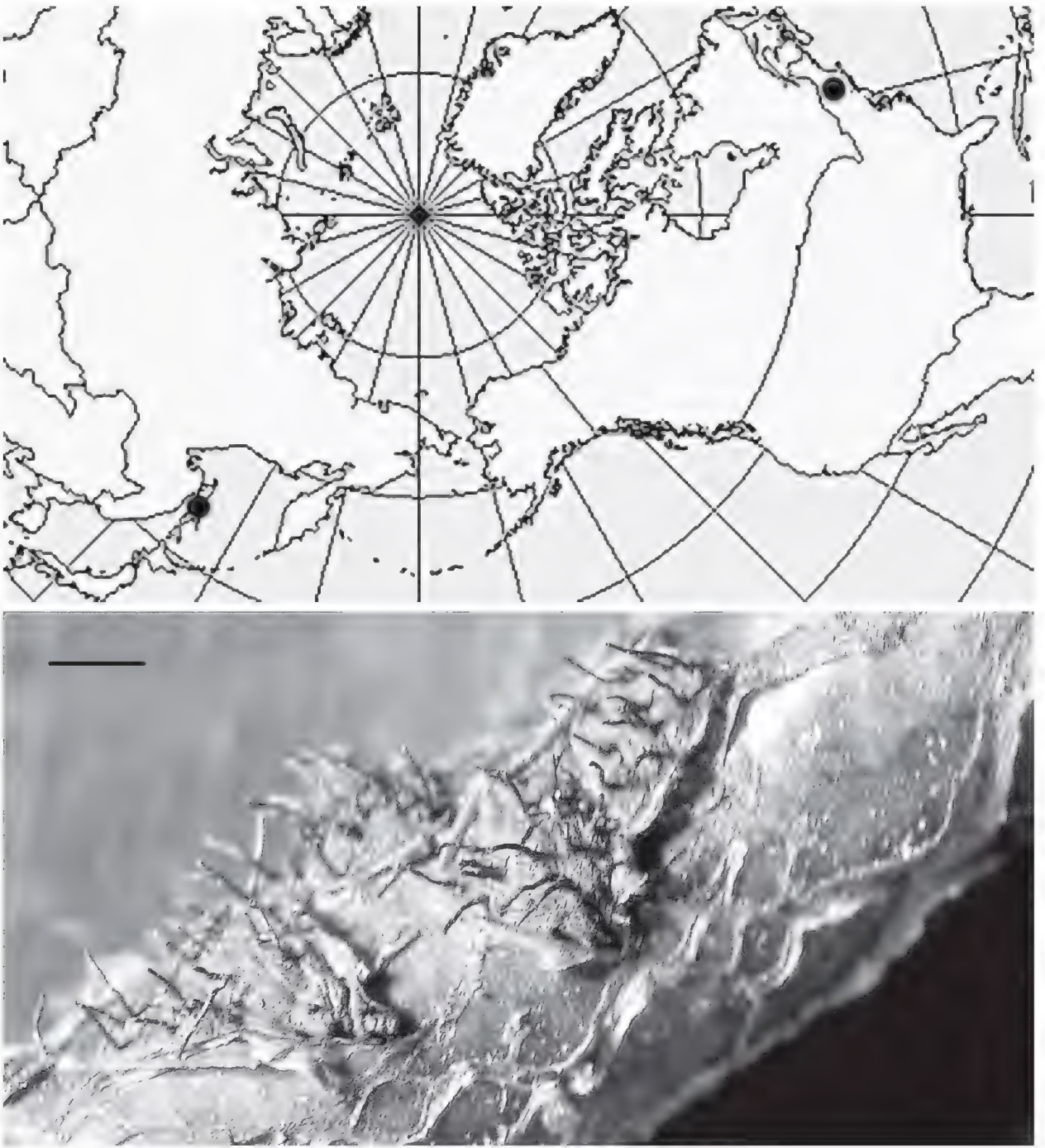


FIG. 7. Known localities for *Diaporthella platasca*. Scale bar = 1.25 mm

literature (Karsten 1873, Klebahn 1918, Monod 1983) indicate ascospores 8–10  $\mu\text{m}$  long for *Apiognomonia alniella*, shorter than Barr's dimensions (10–16  $\mu\text{m}$ ) for her eastern North American material. This suggests that another species occurs in North America that might also be found in eastern Russia at the same latitudes.

We examined one specimen listed by Barr (Quebec: Manitou Gorge, 12 June 1955, R.T. Wilce) that appears exactly the same as specimens from Magadan region, with similar perithecia on dead leaves and same sized ascospores. Even the host leaves looked like those of *Alnus fruticosa*, sometimes referred to *Duschekia* and which supports an array of host-specific pyrenomycetes not



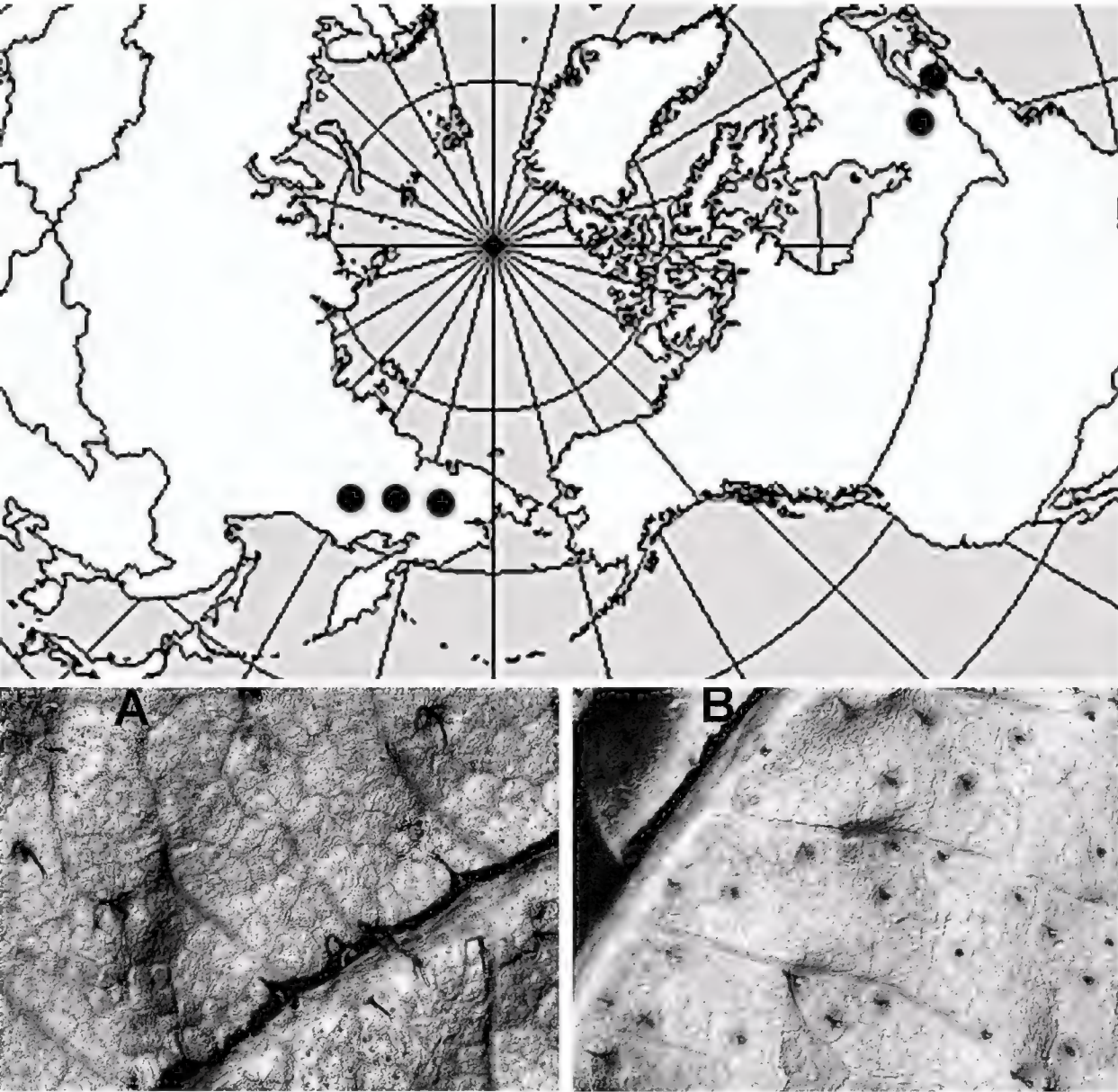


FIG. 8. Approximate biogeographical distribution of *Apiognomonina duschekiae*. The North American are from Barr (1978). In northeastern Russia, this species is rather common and found in many localities within the Magadan Region (vicinities of Kulu and Susuman; the Bol'shoy Anuy, Ilirneyveem, Kegali, Machvavaam, and Yasachnaya river basins; Lake Nizhny Ilirney) The same species should occur on dead leaves of *Duschekia fruticosa* in Yakutia (Shkarupa 1980), although we could not locate the specimen. A - elongated perithecial necks of *A. duschekiae* erumpent from leaf tissue. B - perithecial necks of *A. alniella* (from a specimen collected on *Alnus hirsuta* on the Kamchatka Peninsula).

found on true *Alnus* spp. (a kind of a substrate vicariance). For this reason, we describe below a new species of *Apiognomonina* (*A. duschekiae*), which seems to have a Grayan distribution (FIG. 8).

A similar situation can be observed in a specimen from the Magadan region identified as *Pleuroceras pleurostylum* (Auersw.) M.E. Barr following Barr's concept (Barr 1978, Vasilyeva 1987). Ascospores in the specimen averaged 50–70  $\mu\text{m}$  long, corresponding with Barr's measurements of (35–)40–63 (–72)  $\mu\text{m}$  long. However, Monod (1983) described *P. pleurostylum* occurring

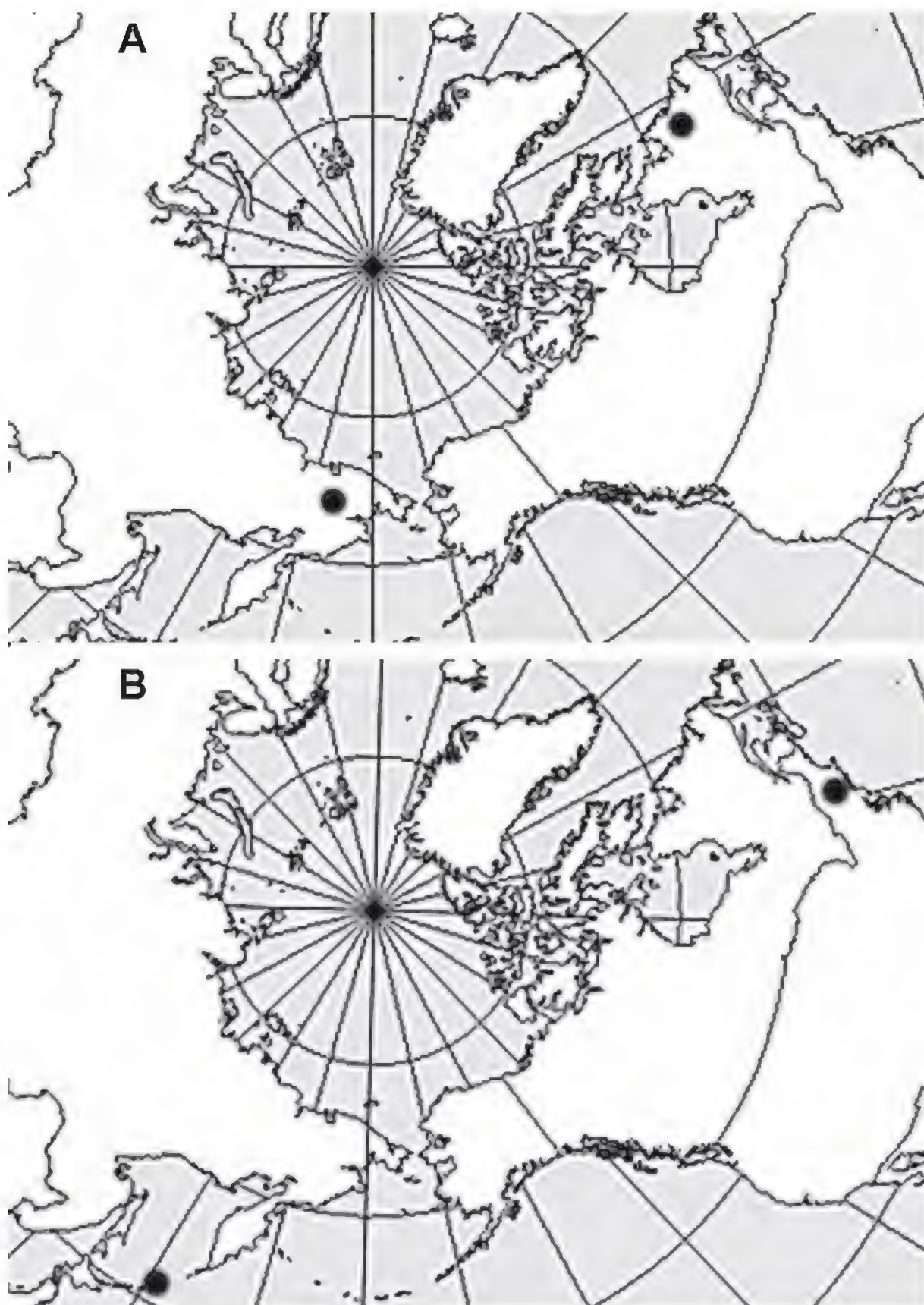


FIG. 9. Approximate biogeographical distributions:  
A - *Pleuroceras labradorensis*, B - *Gnomonia mirabilis*

in Europe as having ascospores 33–45  $\mu\text{m}$  long, whereas the specimen from Labrador cited by Barr with longer (55–67  $\mu\text{m}$ ) ascospores served as the type for his new species *Pleuroceras labradorensis* M. Monod. The collection from



the Magadan region better fits the description of *P. labradorensis*, which appears to have disjunctive distribution in northeastern Asia and northeastern North America (FIG. 9A).

The same situation appears to be the case for *Gnomonia mirabilis* (Peck) M. Monod (FIG. 9B), which occurs on dead leaves of *Betula* spp. This fungus known from North America (New York: Barr 1978) was later found in eastern Asia (Kunashir Island: Vasilyeva 1998). Barr (1978) considered this taxon only a variety of *Plagiostoma campylostyla* (Auersw.) M.E. Barr, but *Gnomonia mirabilis* has appreciably longer ascospores (27.5–37.5 µm versus 18–27 µm in *P. campylostyla*). As this ‘varietal’ difference is surely larger than the difference Barr cited in her key to differentiate *Gnomonia fasciculata* Fuckel from *G. rhoicola* M.E. Barr (ascospores 11–15 and 13–16.5 µm long, respectively), it would seem appropriate also to distinguish *G. mirabilis* and *G. campylostyla* Auersw. at the species level. Monod (1983) listed three additional localities (Michigan, Ontario, Quebec) in eastern North America for *G. mirabilis*. The fungus probably occurs there, but Monod’s species description cites an ascospore length of 21–32 µm, which would not distinguish it from *G. campylostyla*. Monod does not compare *G. mirabilis* with any other species of that genus in his key.

It is particularly noteworthy how often diaporthalean fungi display a Grayan disjunction. Recently, *Melanconis carpinigera* (Ellis & M.A. Curtis) Petr. was reported from eastern Russia (the Vladivostok vicinity), a species known previously from eastern North America (Wehmeyer 1941, as *M. chrysostroma* var. *ellisii* (Rehm) Wehm.) (FIG. 10) and the third species recorded from *Carpinus cordata* (along with *Fracchiæa callista* and “*Diatrypella informis*”, discussed above) with such a distribution.

Testing for a Grayan distribution pattern may be useful for species already known from eastern North America when the same species is found in eastern Asia, particularly a taxonomic change might be indicated. For example, *Hypoxylon lividipigmentum* F. San Martín et al. was described from Mexico as having a teleomorph that is almost identical to *H. lividicolor* Y.M. Ju & J.D. Rogers known from Taiwan, except for the fact that the stromata of the former are thinner. Two species collected at almost the same latitudes (near the Northern tropics) in eastern North America and eastern Asia certainly warrant careful comparison. The senior author found a similar fungus in Texas (within the Big Thicket National Preserve), and there were reasons to identify it as *Hypoxylon lividipigmentum*, described from neighboring Mexico (the state of Quintana Roo), since southern Texas appears to share numerous species of pyrenomycetous fungi with Mexico. However, the stromata in the Texan specimen were rather thick, and J.D. Rogers (pers. comm.) was inclined to consider it to represent the Taiwanese *H. lividicolor*. The most probable conclusion is that the Taiwanese, Mexican, and Texan specimens belong to the same species being variable



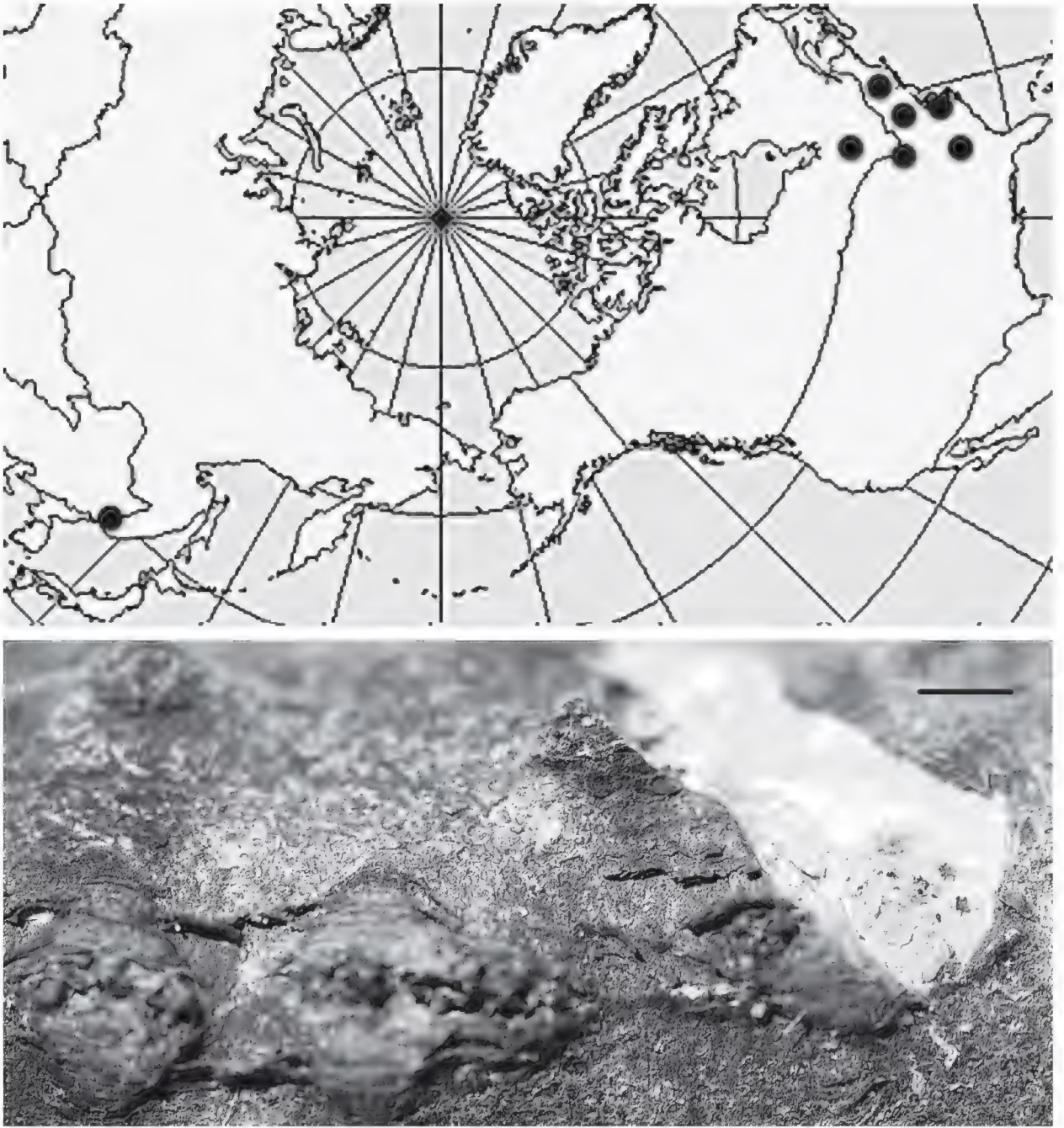


FIG. 10. Approximate biogeographical distribution of *Melanconis carpinigera*. North American localities as cited by Wehmeyer (1941: Michigan, New York, Pennsylvania, Ontario) as collected by the senior author (Maryland—BPI 843491; Tennessee—878343). Scale bar = 1.4 mm

in stromatal thickness, and this species displays a familiar disjunction in its distribution. *Hypoxylon lividipigmentum* and *H. lividicolor* are currently treated as separate species, since one of them has a *Nodulisporium*-like conidiogenous structure, whereas the other has a conidiogenous structure that is *Sporothrix*-like (Ju & Rogers 1996). However, both *Nodulisporium*- and *Sporothrix*-like have been reported to occur within the same species (e.g., *Hypoxylon macrosporum* P. Karst.). Otherwise, *Hypoxylon lividipigmentum* and *H. lividicolor* represent a vicariance pattern in Grayan distribution (FIG. 11), and the specimen from Texas belongs to *H. lividipigmentum* despite its rather thick stromata.



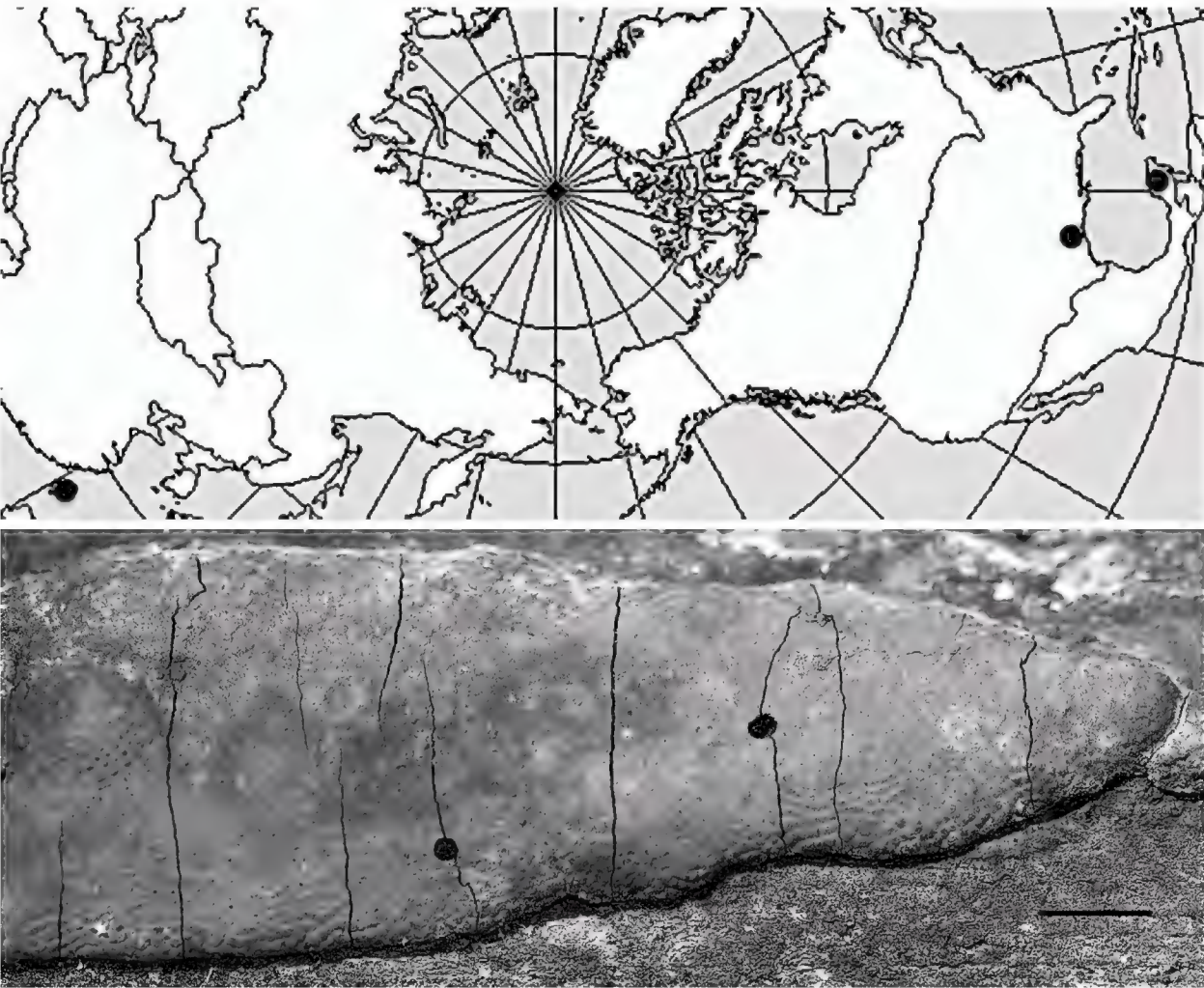


FIG. 11. Approximate biogeographical distribution of *Hypoxylon lividicolor* (Taiwan) and *H. lividipigmentum* (Mexico, Texas). Stroma: VLA P-2450 (Texas). Scale bar = 0.6 mm

Vicariance pattern

The vicariance pattern in Grayan distribution observed for some pyrenomycetous fungi is an even more interesting topic for discussion, since several species were described from eastern Russia as counterparts of eastern North American relatives, but only as varieties or synonyms of the latter. This additionally emphasizes an important taxonomic problem associated with estimating of differences in rank, which could be resolved by considering vicariant species pairs in eastern Asia and eastern North America.

One noteworthy example is *Biscogniauxia maritima* Lar.N. Vassiljeva, described as an east-Asian counterpart of the North American *B. atropunctata* (Schwein.) Pouzar (FIG. 12). In their disjunct regions, both species are restricted to *Quercus* spp. but differ considerably in ascospore size ( $13.2\text{--}16 \times 6.6\text{--}8 \mu\text{m}$  versus  $24\text{--}33 \times 11\text{--}16 \mu\text{m}$ ). Although *B. maritima* was later reduced to a variety of *B. atropunctata* (Ju et al. 1998), the vicariance pattern remains.

Nevertheless, the status of a taxon as a species or a variety is of importance, and the rank is determined after a careful consideration of differences that



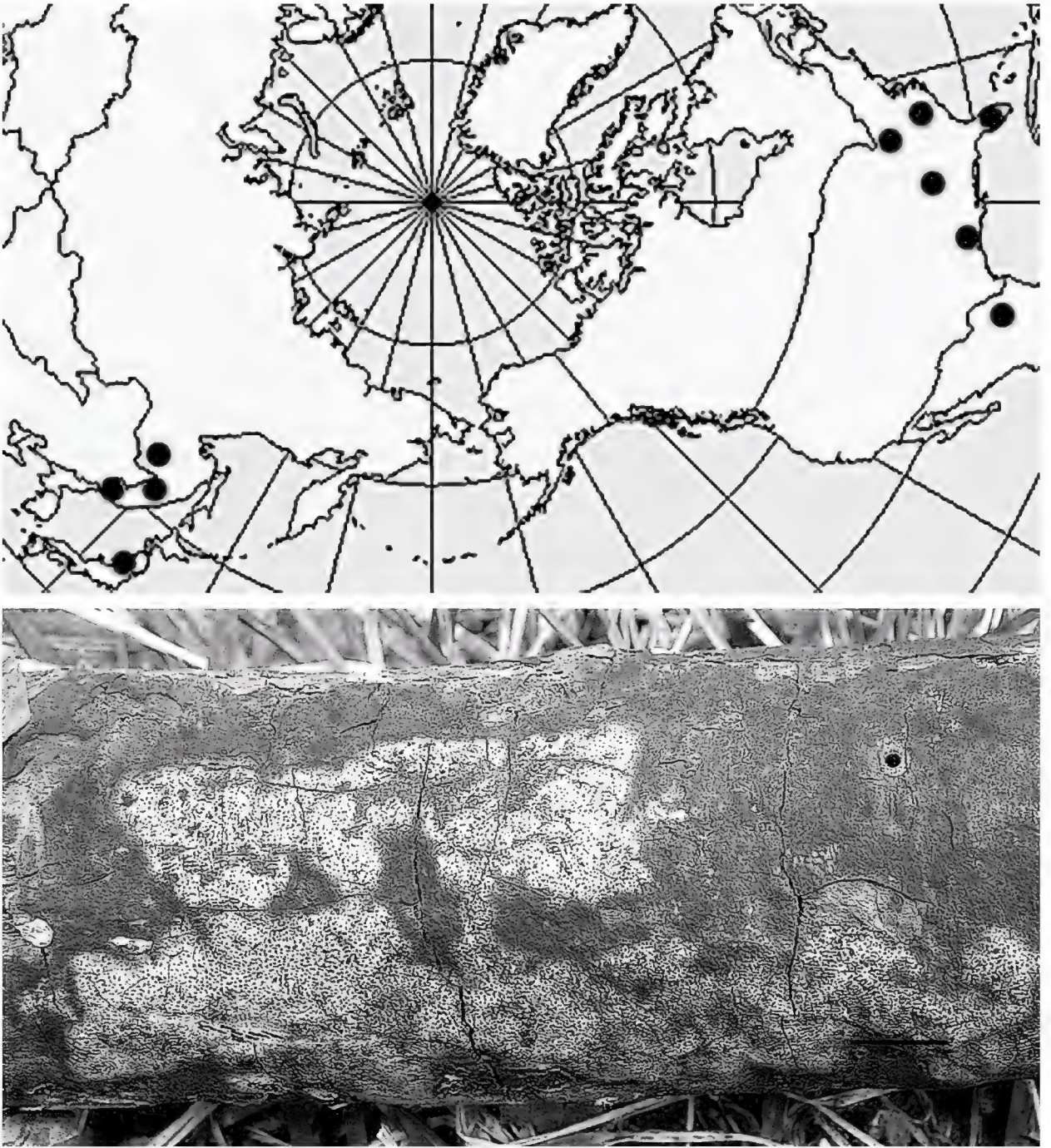


FIG. 12. Approximate biogeographical distributions of *Biscogniauxia maritima* and *B. atropunctata*. North American localities from Mexico (Nuevo León state)] and the USA (Florida, North Carolina, Ohio) are based on Ju et al. (1998) and collections by the senior author in Arkansas (Buffalo National River) and Texas (Big Thicket National Preserve). The species was also found in Tennessee (the Great Smoky Mountains National Park). All eastern Asian collections were obtained by L. Vasilyeva. Scale bar = 0.6 mm

exist within a particular genus or (sometimes) among several closely related genera. An examination of the key to *Biscogniauxia* taxa by Ju et al. (1998) reveals immediately that many taxa differ only in ascospore size (steps 3, 6, 9, 33–34, 36, 40), implying that these taxa are similar in other features. Using these examples (i.e., taxa distinguished at the steps indicated), we arranged the species and varieties on the basis of the average lengths of ascospores (TABLE 1), with each table row presenting a set of closely related taxa.

TABLE 1. Arrangement of some *Biscogniauxia* taxa in accordance with average ascospore length (data from Ju et al. 1998: Key to *Biscogniauxia*). Each table row presents a set of closely related taxa.

ASCOSPORE LENGTH			
10–13 µm	13–17 µm	18–22 µm	22–30 µm
		<i>B. weldenii</i> var. <i>microspora</i>	<i>B. weldenii</i>
	<i>B. nothofagi</i>		<i>B. pithodes</i>
	<i>B. philippinensis</i> var. <i>microspora</i>		<i>B. philippinensis</i>
<i>B. uniapiculata</i>	<i>B. uniapiculata</i> var. <i>macrospora</i>	<i>B. divergens</i>	
	<i>B. maritima</i>	<i>B. atropunctata</i> var. <i>intermedia</i>	<i>B. atropunctata</i>
<i>B. citriformis</i>	<i>B. citriformis</i> var. <i>macrospora</i>		
<i>B. nummularia</i>		<i>B. bartholomaei</i>	
	<i>B. mediterranea</i> var. <i>microspora</i>	<i>B. mediterranea</i>	

Consideration of the information in TABLE 1 shows, for example, that the same difference exists between *Biscogniauxia nothofagi* Whalley et al. and *B. pithodes* (Berk. & Broome) Whalley & Læssøe, *B. philippinensis* (Ricker) Whalley & Læssøe and *B. philippinensis* var. *microspora* Y.M. Ju & J.D. Rogers, and *B. atropunctata* and *B. atropunctata* var. *maritima* (Lar.N. Vassiljeva) Y.M. Ju & J.D. Rogers. Yet the taxa in the first pair are treated as different species, whereas the others are regarded only as varieties. That is taxonomically inconsistent, since the same character difference should not be used at two ranks in the same genus, and if ‘varieties’ within *Biscogniauxia* display their own biogeographical patterns as do the varieties *atropunctata* and *maritima* of *B. atropunctata* in eastern Asia and eastern North America (FIG. 12), they probably deserve recognition at the species level, as *B. nothofagi* and *B. pithodes* are recognized.

*Biscogniauxia mediterranea* (De Not.) Kuntze and *B. mediterranea* var. *microspora* display a substrate vicariance, with the autonymous variety occurring only on *Quercus* spp. and var. *microspora* not occurring on *Quercus* but seemingly preferring *Alnus* spp. The latter was found several times on *Alnus* in British Columbia and California (Ju et al. 1998), while are collections from eastern Russia (Khanka Nature Reserve) on *Corylus heterophylla* Fisch. ex Trautv., also in the *Betulaceae*. We regard *B. mediterranea* var. *microspora* as a separate species, for which we propose the name *Biscogniauxia alnophila* below.

Another example of a species with a vicariance pattern is *Hypoxylon ulmophilum* Lar.N. Vassiljeva, common on dead branches of *Ulmus* spp. in the Russian Far East. Vasilyeva (1998) described it as having glomerate stromata

TABLE 2. Arrangement of some *Hypoxylon* taxa in accordance with average ascospore length (data from Ju & Rogers 1996: Key to *Hypoxylon*). Each table row presents a set of closely related taxa.

ASCOSPORE LENGTH			
7–11 µm	11–15 µm	15–22 µm	22–26 µm
<i>H. howeanum</i>	<i>H. fragiforme</i>		
<i>H. aeruginosum</i>	<i>H. aeruginosum</i> var. <i>macrosporum</i>		
<i>H. monticulosum</i>	<i>H. rubigineoareolatum</i>		
<i>H. carneum</i>			<i>H. vogesiacum</i>
	<i>H. notatum</i>	<i>H. ulmophilum</i>	
<i>H. investiens</i>		<i>H. subcorticeum</i>	
	<i>H. ferrugineum</i>	<i>H. diatrypeoides</i>	
<i>H. annulatum</i>		<i>H. thouarsianum</i>	<i>H. thouarsianum</i> var. <i>macrosporum</i>
<i>H. leptascum</i>		<i>H. leptascum</i> var. <i>macrosporum</i>	

similar to those found in *Hypoxylon notatum* Berk. & M.A. Curtis but differing in larger ascospores (16.5–21 µm versus 12–15 µm long). Ju et al. (2004) rejected the new species as conspecific with *H. notatum*, but Stadler et al. (2008) later supported it as an independent taxon.

The larger ascospores, a different substrate preference, and the apparent biogeographical pattern suggested a different species. However, once again, the question could be asked as to whether it is possible to rely only upon a single morphological difference, such as the ascospore size. As with ascospore size in *Biscogniauxia*, repetitive average lengths also exist within *Hypoxylon*, where also species (and varieties) appear to differ only in ascospore size. TABLE 2 compares size differences (steps 7, 12, 31, 33, p. 58–62, etc.) in the key by Ju & Rogers (1996). One can see that the table for *Hypoxylon* contains fewer varieties when compared with the table for *Biscogniauxia*. In other words, average ascospore size (comparable in both TABLE 1 and TABLE 2) serves to delimit species in many instances, although using the same difference to delineate both species and varieties is inconsistently applied. If a number of species differ only in ascospore size, then, following simple taxonomic logic, there is justification for recognizing *Hypoxylon notatum* and *H. ulmophilum* as different species.

The concept of *Hypoxylon notatum* in the monograph by Miller (1961) is rather narrow, indicating that it occurs primarily on *Quercus* spp. in the eastern United States (FIG. 13). As such, *H. notatum* represents a counterpart to *H. ulmophilum* in the vicariance pattern under discussion. Later, the concept of *H. notatum* was widened to include some species described from Brazil and Paraguay, as well as specimens from tropical China and Taiwan (Ju & Rogers 1996). In this broader sense, what is currently recognized as *H. notatum* might



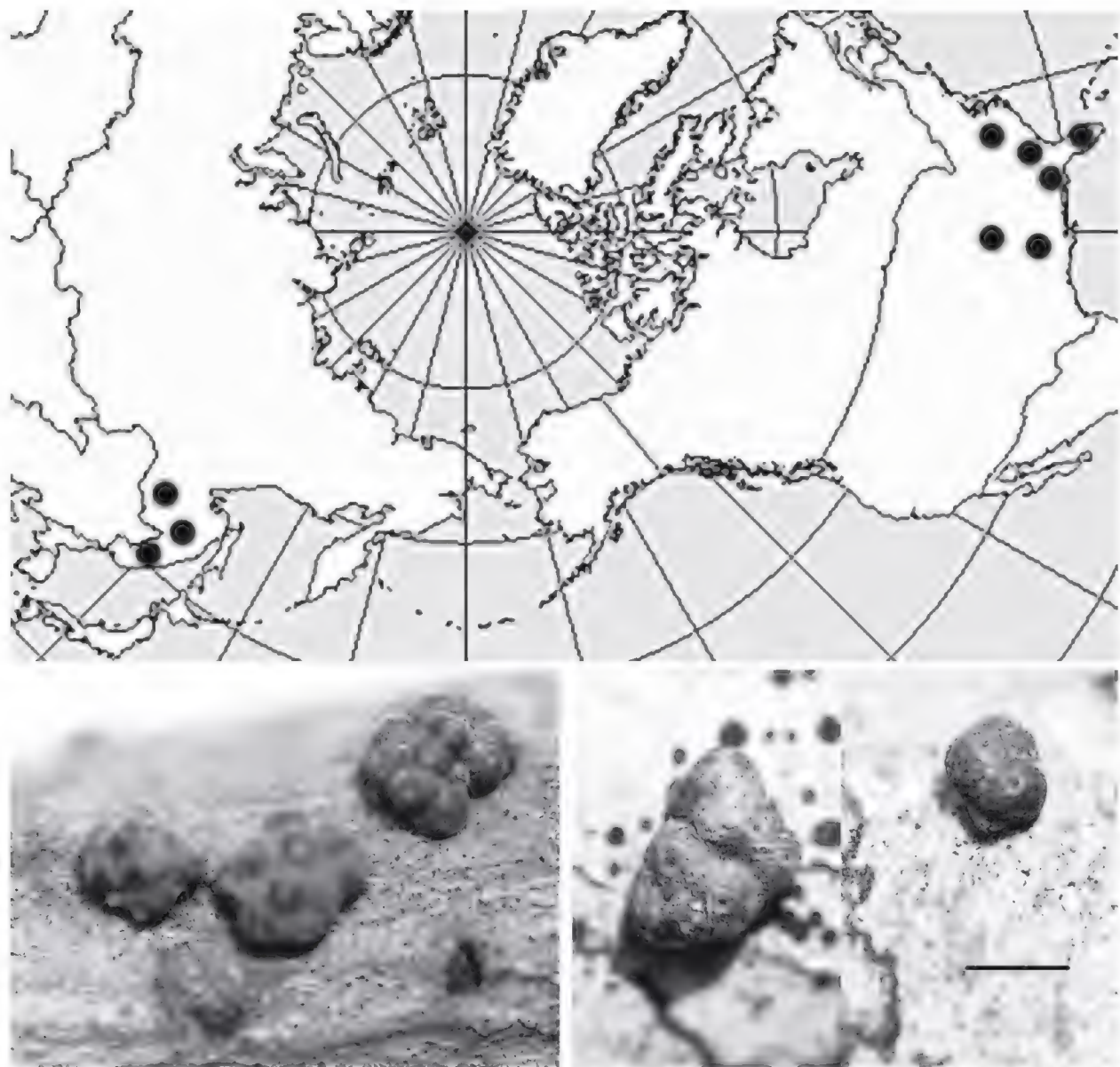


FIG. 13. Approximate biogeographical distribution of *Hypoxylon ulmophilum* (left) and *H. notatum* (right). The North American localities of *H. notatum* are those cited by Miller (1961). The *H. ulmophilum* collections in eastern Asia were obtained by L. Vasilyeva. The latter species was also found in South Korea (Gangwon province, Mt. Odaesan, 20 Sep 2006, VLA P-1651). A. Stromata of *H. ulmophilum*. B. Stromata of *H. notatum*: the widely opening mouth-like ostioles are diagnostic (cf. also Miller 1961, FIG. 7). Scale bar = 1.1 cm

represent a species complex in need of reconsideration. Some support for this view was provided by a specimen from Texas (Big Thicket National Preserve) that is very similar to *Hypoxylon notatum* as illustrated by Miller (1961: FIG. 6–7), but the KOH-extractable stromatal pigments of the Texan specimen are orange in contrast to “pure yellow with greenish yellow tone” reported for *H. notatum* by Ju & Rogers (1996). The latter pigment type was confirmed only for the Taiwanese specimen (Stadler et al. 2008), whereas material from Argentina identified as *H. notatum* had light chestnut pigments (Hladki & Romero 2006). The specimens from the USA studied by Stadler et al. (2008) had a more or less dilute umber pigment in KOH.

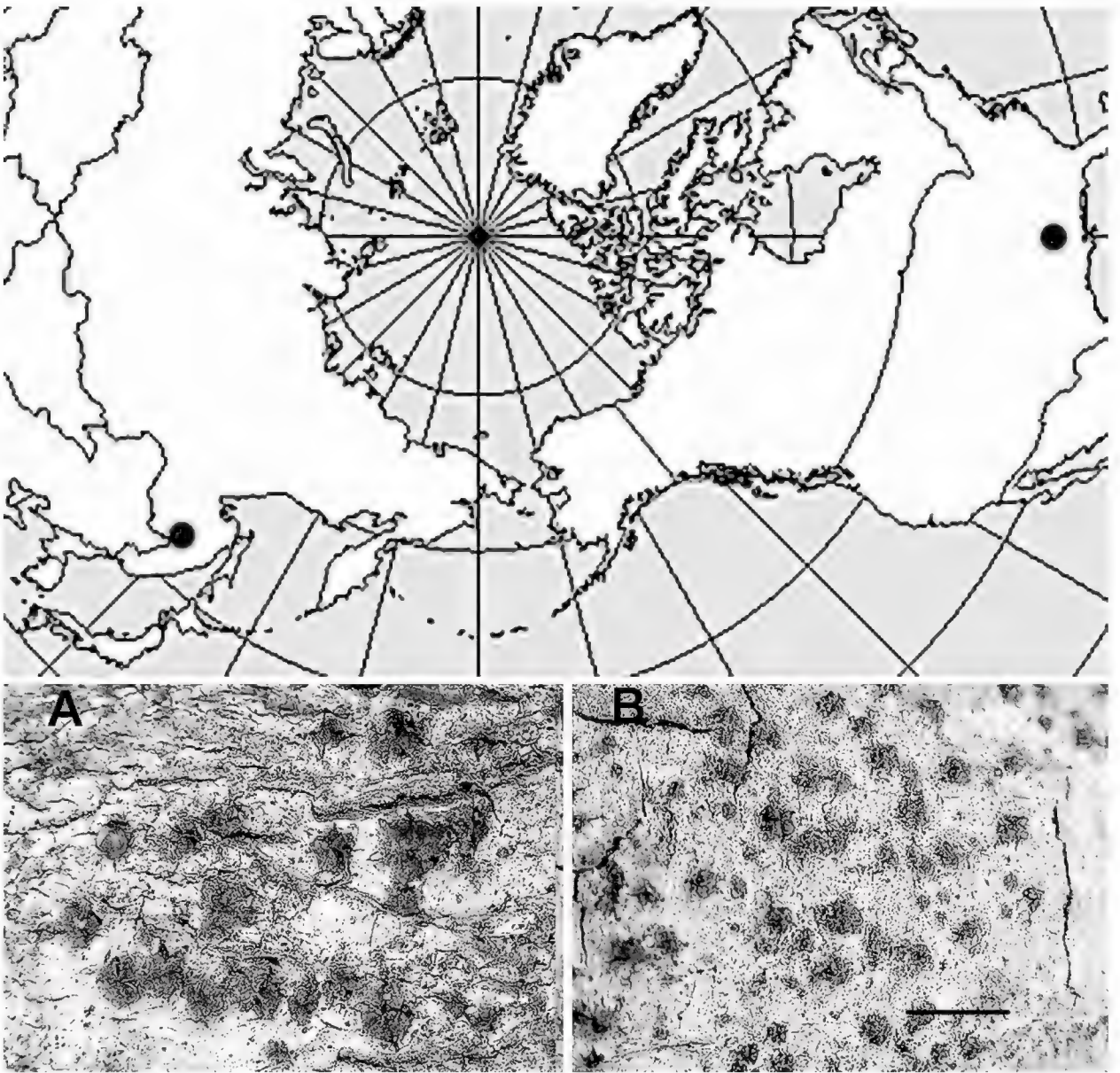


FIG. 14. Known localities for *Cryptovalsaria rossica* and *C. americana* in eastern Russia (the Khabarovsk vicinity) and the USA (Ouachita Mountains, Arkansas). A - Ostioles of *C. rossica* on bark. B - Smaller ostioles of *C. americana* on bark. Scale bar = 2.5 mm

There is no reason to extend this paper by listing additional pairs of pyrenomycetous species that display the vicariance pattern within the Grayan distribution. It is sufficient to mention a very curious case involving closely related species in eastern Asia and southeastern North America parasitizing the same kinds of host trees. These are *Cryptovalsaria rossica* Lar.N. Vassiljeva & S.L. Stephenson and *C. americana* Lar.N. Vassiljeva & S.L. Stephenson, found on living trees of *Alnus* spp. in eastern Russia and Arkansas (FIG. 15) within a time period of six years (Vasilyeva & Stephenson 2007). The situation with these two species is very similar to that with two other ascomycetous species (Whetzel & Wolf 1945), namely *Ciboria shiraiana* (Henn.) Whetzel and *C. carunculoides* (Siegler & Jenkins) Whetzel, parasitizing the fruits of *Morus* spp. in eastern Asia (Japan, South Korea, China, southeastern Russia) and the



southeastern United States (Alabama, Arkansas, Georgia, Florida, Louisiana, Mississippi, North Carolina, South Carolina, Texas).

## Taxonomy

***Apiognomonia duschekiae*** Lar.N. Vassiljeva & S.L. Stephenson, **sp. nov.**

MYCOBANK MB 518664

*Perithecia singula, immersa, ut plurimum ad nervi sparsa, sed frequenter ad laminae quoque dispersa, nigra, globosa, 250–300 µm diametro, cum rostri centrali, tenui, recti vel curvati, ad 300–500 µm longi, hypophylli vel epiphylli. Asci numerosi, ellipsoidei, octospori, 50–66 × 9–12 µm. Ascosporae hyalinae, ellipsoideae vel fusioideae, prope basim uniseptatae, ad septum non constrictae, 12–14(–16) × 5–6.6 µm.*

**HOLOTYPE:** Russia, Magadan Region, Susuman vicinity, on dead leaves of *Duschekia fruticosa* (Rupr.) Pouzar (*Betulaceae*), 19.VII.1974, L. Vasilyeva, VLA P-1093.

*Perithecia* solitary, immersed in leaf tissue, most often along veins, black, spherical, 250–300 µm diam., with central, thin, straight or curved necks up to 300–500 µm long, emerging from the lower or upper leaf surface. Asci numerous, ellipsoid, 8-sporous, 50–66 × 9–12 µm. Ascospores hyaline, ellipsoid or fusiform, septate near basis, not constricted, 12–14(–16) × 5–6.6 µm.

**ADDITIONAL SPECIMENS EXAMINED:** All specimens were collected from dead leaves of *Duschekia fruticosa* by L. Vasilyeva and are deposited in VLA: MAGADAN REGION, Ygodnino District, basin of the river Yasachnaya, 19.VII.1975, P-893; Severo-Evensk District, basin of the river Kegali, 6.VIII.1976, P-894; Bilibino District, basin of the river Ulyashka, 3.VII. 1976, P-892; basin of the river Machvavaam, 13.VII.1977, P-887; basin of the river Bol'shoy Anuy, 25.VII.1980, P-890; basin of the river Ilirney, 13.VIII.1980, P-891; lake Nizhny Ilirney, 21.VIII.1980, P-888; District Ten'kinsky, Kulu vicinity, 12.IX.1975, P-895.

***Biscogniauxia alnophila*** Lar.N. Vassiljeva & S.L. Stephenson, **nom. nov.**

MYCOBANK MB 518754

= *Hypoxylon mediterraneum* var. *microsporum* J.H. Mill., Monogr. of the World Species of *Hypoxylon*: 117 (1961).

= *Biscogniauxia mediterranea* var. *microspora* (J.H. Mill.) Y.M. Ju & J.D. Rogers, Mycotaxon 66: 42 (1998).

**DESCRIPTION**—Miller (1961: 117), Ju et al. (1998: 42).

**SPECIMENS EXAMINED:** RUSSIA, PRIMORSKY TERRITORY: Khanka Nature Reserve, on dead branches of *Corylus heterophylla*, 18 Jun 2003, L. Vasilyeva, VLA P-1858.

***Nemania sphaeriosstoma*** (Schwein.) Lar.N. Vassiljeva & S.L. Stephenson, **comb. nov.**

MYCOBANK MB 518690

= *Sphaeria sphaeriosstoma* Schwein., Trans. Amer. Philos. Soc., n. ser. 4: 193 (1832).

= *Hypoxylon sphaeriosstomum* (Schwein.) Sacc., Syll. Fung. 1 : 392 (1882).

**DESCRIPTION**—Miller (1961: 67; FIGS. 100, 128).

**SPECIMENS EXAMINED:** RUSSIA, PRIMORSKY TERRITORY: Lazovsky Nature Reserve, on wood, 2 Aug 1986, L. Vasilyeva, VLA P-380; Ussuriysky Nature Reserve, on wood, 18 Sep 1996, L. Vasilyeva, VLA P-379.

COMMENTS—Superficially, the stromata in the Asian specimens of *Nemania sphaerostoma* (FIG. 5) and in Miller's photograph (1961: FIG. 100) are similar to both *Euepixylon udum* (Pers.) Læssøe & Spooner and *Nemania confluens* (Tode) Læssøe & Spooner (Granmo et al. 1999: Figs. 17, 42), and the distribution of the two latter species among different genera was made on the basis of *Euepixylon udum* having ascospores with elliptic, poroid germ slit, whereas *Nemania confluens* is characterized by ascospores with a narrow, long germ slit. However, this difference is hardly of generic importance, since the size of germ slit (which in some instances is seemingly lacking) varies within many genera of the *Xylariaceae*, within which this difference is usually used to distinguish species in such genera as *Hypoxylon* (Ju & Rogers 1996), *Biscogniauxia* (Ju et al. 1998) and *Nemania* (Ju & Rogers 2002).

When reinstated, the genus *Euepixylon* was distinguished from *Nemania* on the basis of “a short poroid germ locus, a very short ascus stipe, and a broad, discoid apical apparatus” (Læssøe & Spooner 1993: 41), but the authors themselves expressed doubts that this genus would survive in the long run. Later, the name *Euepixylon* was said to be invalid (Eriksson & Hawksworth 1997), so the genus *Nemania* is more suitable for *Hypoxylon sphaerostomum* on the basis of both the logics of taxonomic comparison as well as nomenclatural rules.

### Acknowledgments

The authors are grateful to Dr. R.H. Petersen of the University of Tennessee and H.H. Burdsall of Fungal and Decay Diagnostics, LLC for serving as presubmission reviewers and for providing helpful comments and suggestions. One of the earlier drafts of this manuscript was kindly looked over by Dr. A.Y. Rossman of the Agricultural Research Service and her comments are greatly appreciated.

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## MYCOTAXON

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**First record of *Phlebia incarnata*  
from the southern hemisphere****MAURO C. WESTPHALEN, MATEUS A. RECK  
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**Abstract** — During a survey of xylophilous fungi in the municipality of São Francisco de Paula, in southern Brazil, *Phlebia incarnata*, a species never before recorded for South America, was found. *Phlebia incarnata* has a pileate basidiome with vivid pink coloration, a hymenophore with folds, a monomitric hyphal system, and cylindrical basidiospores. In this work, *P. incarnata* is compared with related species; a full description and illustrations are provided.

**Key words** — *Meruliaceae*, *Merulius*, mycodiversity, neotropics

**Introduction**

The genera *Merulius* Fr. and *Phlebia* Fr. were described by Fries in 1821. Since then, they have both been placed in family *Meruliaceae* P. Karst. (Kirk et al. 2008) and are mainly differentiated by the habit (reflexed to dimidiate in the former and resupinate to effused in the latter). Fries (1821) included 10 species in the genus *Merulius* and later divided it into two sections according to the pigmentation of the basidiospores (Fries 1838). Patouillard (1887) transferred the *Merulius* species with colored spores to the genus *Gyrophora* Pat., and later Karsten (1889) divided *Merulius* into four distinct genera: *Merulius*, *Plicatura* Peck, *Gyrophora*, and *Serpula* (Pers.) Gray. Over the years, further work including these genera has been published (Patouillard 1900, Donk 1964, Parmasto 1968), but none satisfactorily distinguished *Merulius* and *Phlebia*. They only agreed with Karsten's (1889) idea that they were related and difficult to discern due to morphological similarities. Ginns (1975), based on morphological and cultural characters, proposed a new segregation of the species of *Merulius* s.l., keeping

**NOTE** —MYCOTAXON prepared this PDF with color plates for the author. The original print version was published with halftone (grayscale) plates.

only two species in *Merulius* s.s., *M. tremellosus* Schrad., and *M. incarnatus* Schwein. However, Nakasone & Burdsall (1984), who morphologically and culturally compared the type species of *Merulius* and *Phlebia* (*M. tremellosus* and *P. radiata* Fr.), considered that the differences presented (based on basidiome habit, configuration of the hymenophore, and presence of cystidia and asexual spores in culture) were not sufficient to separate them into two different genera. Consequently they considered *Merulius* a synonym of *Phlebia*.

Using RFLP analysis of 18S rRNA gene fragment and ITS region, Dresler-Nurmi et al. (1999) demonstrated that *Phlebia tremellosa* (Schrad.) Nakasone & Burds. grouped together with *P. ochraceofulva* (Bourdot & Galzin) Donk, *P. centrifuga* P. Karst., and *P. radiata*. Subsequent phylogenetic analysis of the sequences 5.8S, ITS2, and LSU nuclear rDNA by Larsson et al. (2004) showed that *P. tremellosa* is closely related to *P. rufa* (Pers.) M.P. Christ., *P. radiata*, and *P. lindtneri* (Pilát) Parmasto. As *P. tremellosa* and *P. incarnata* are very similar morphologically, and the former groups in the same clade with other *Phlebia* species, it is likely that both belong to this genus instead of *Merulius*, thus supporting the conclusions of Nakasone & Burdsall (1984).

*Phlebia* is characterized by effuse to effuse-reflex or dimidiate basidiomata with cartilaginous to subgelatinous or ceraceous consistency. Hymenial surfaces can be smooth, tuberculate, odontoid, phlebioid, or merulioid. A monomitric hyphal system and smooth, thin-walled and non-amyloid basidiospores characterize the genus microscopically (Nakasone & Burdsall 1984, Maekawa 1993).

### Materials and methods

Specimens were collected in July and September 2009, in the municipality of São Francisco de Paula, Rio Grande do Sul, Brazil. This region is characterized by presenting subtropical vegetation with the presence of the coniferous tree *Araucaria angustifolia* (Bertol.) Kuntze (*Araucariaceae* Henkel & W. Hochst.). The climate in the region is humid subtropical of the Cfb type, according to the Köppen Climate Classification (Moreno 1961).

After the macromorphological analysis, the specimens were dried at room temperature. For microscopy, freehand basidiome sections were mounted in a drop of 5% KOH solution and 1% phloxine solution. Microstructures were drawn aided by a camera lucida. The abbreviations and codes for the measurements are modified from Coelho (2005), where  $Lm \times Wm$  = means of length and width,  $Q$  = range of length/width ratios,  $Qm$  = length/width mean, and  $n = x/y$  ( $x$  = number of measurements from a given number ( $y$ ) of specimens). The codes used for colors follow Kornerup & Wanscher (1978). The collected specimens are kept at the ICN herbarium (UFRGS).

## Taxonomy

*Phlebia incarnata* (Schwein.) Nakasone & Burds., Mycotaxon 21: 245, 1984

FIGS 1–5

SPECIMEN EXAMINED: BRAZIL. Rio Grande do Sul, municipality of São Francisco de Paula, FLONA, 03.VII.2009, leg. G. Seger 1028 (ICN 154337); 19.IX.2009, leg. G. Seger 1029 (ICN 154388).

BASIDIOMATA annual, pileate, sessile to dimidiate, sometimes slightly effused-reflexed, often imbricate, spongy when fresh becoming hard upon drying, pileus conchate; upper surface tomentose, pinkish to reddish (11A4–12A7) when fresh and pinkish white to reddish blond (7A2–5C3) after dried; margin fimbriate, vivid red (11A8); hymenial surface white (11A1) when fresh, drying dull red (9C4–10B4), folds 0.5–1.0 mm deep, radiating, continuous to the margin, side branches anastomosing forming cavities resembling a pore surface (1–2/mm); context up to 2.0 mm thick, duplex, upper layer loose and spongy, concolorous with the upper surface, lower layer waxy and dense, brownish red (10D6) to dull red (11C4).

HYPHAL SYSTEM monomitic, generative hyphae with clamp connections, 2.0–5.0  $\mu\text{m}$  diam., thin to slightly thick-walled, with wide lumen, amorphous granules present in contextual hyphae; cystidia lacking. Basidia clavate, 4-sterigmate; basidiospores subcylindrical to cylindrical, slightly bent, hyaline, smooth, thin walled, frequently with two oil drops,  $4.5\text{--}5.5 \times 2.0\text{--}2.5 \mu\text{m}$ ,  $\text{Lm} \times \text{Wm} = 5.08 \times 2.10$ ,  $Q = 2.0\text{--}2.75$ ,  $Qm = 2.43$ ,  $n = 30/1$ .

CULTURE DESCRIPTION: See Ginns (1975)

SUBSTRATA: On fallen logs of an unknown angiosperm.

DISTRIBUTION: Previously recorded from United States, Mexico (Ginns 1975), and Costa Rica (Halling & Mueller 2006).

ADDITIONAL SPECIMENS EXAMINED: *Phlebia incarnata* – UNITED STATES. North Carolina, Franklin County, Louisburg, 01.II.2003, leg. V. Grand s/n (BPI 844251); Texas,

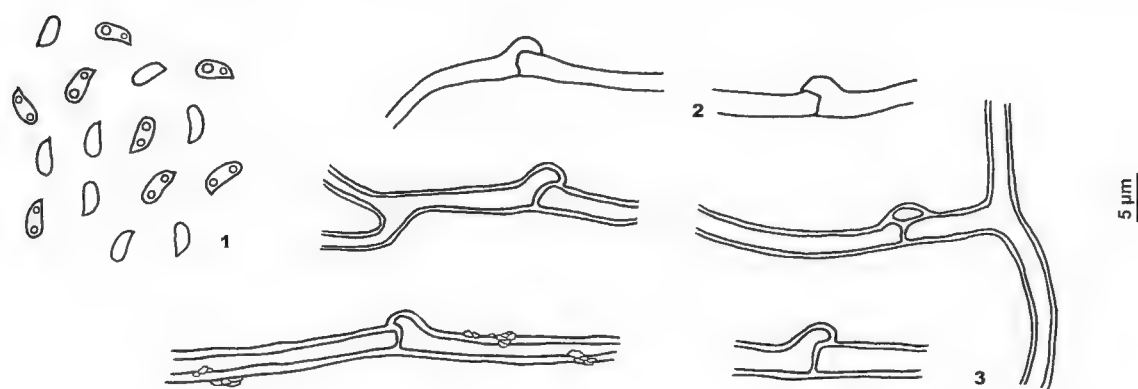


FIG. 1–3. *Phlebia incarnata* (ICN 154337).

1. Basidiospores. 2. Tramal generative hyphae. 3. Contextual generative hyphae.





FIG. 4–5. Basidiome of *Phlebia incarnata*. 4. Pileus surface. 5. Hymenophore. Scale bar = 1 cm.

Hardin County, Big Thicket National Preserve, Jack Gore Baygall Unit, 17.XI.2001, leg. D.P. Lewis 6542 (BPI 841954); Virginia, King George County, 7.XI.1972, leg. K.A. Harrison KHM 13344 (BPI 025617). *Phlebia tremellosa* — BRAZIL. Rio Grande do Sul, municipality of Cambará do Sul, Itaimbezinho, II.1981, leg. R.T. Guerrero s/n (ICN 56048 as *Merulius tremellosus*); municipality of São Francisco de Paula, CPCN Pró-Mata, 29.V.2009, leg. M.C. Westphalen 230/09 (ICN 154339); FLONA, 22.VI.2009, M.C. Westphalen 250/09 (ICN 154338).

REMARKS: *Phlebia incarnata* is easy to recognize due to its vivid reddish-pink color, spongy basidiomata, and folded hymenophore. Our specimens fit the description given by Ginns (1975), differing only in the fresh hymenial surface color, which in our specimens is white, while Ginns describes it as pale pink. Also, the specimens we examined from BPI herbarium usually presented a glabrous upper surface, sometimes with small hairs in restricted areas, while our material presented a tomentose to somewhat velvety upper surface.

*Phlebia tremellosa* is a similar species that also occurs in Brazil (Baltazar & Gibertoni 2009). However it presents a white to pallid pileus surface and the hymenial surface has a translucent pale orange-red coloration, which becomes deep orange-red upon drying. Microscopically, *P. tremellosa* can be differentiated by the allantoid basidiospores ( $4.0\text{--}4.5 \times 1.0\text{--}1.5$ ) and the presence of scattered cystidia imbedded in the hymenium.

According to Ginns (1975), *P. incarnata* frequently grows together with basidiomes of a species of *Stereum* Hill ex Pers. However, in our specimens, we did not observe this association.

This species was previously known only from countries located in the northern hemisphere. Therefore our record represents a significant addition to its biogeography distribution.

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## MYCOTAXON

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**New records of lichenicolous and lichenized fungi  
from Turkey**MEHMET GÖKHAN HALICI<sup>1\*</sup>, ILGAZ AKATA<sup>2</sup> & MUSTAFA KOCAKAYA<sup>3</sup><sup>1</sup>*mghalici@erciyes.edu.tr**Biyoloji Bölümü, Fen Fakültesi, Erciyes Üniversitesi  
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**Abstract** — In the course of studying the lichenicolous and lichenized fungi deposited in the lichen herbarium of Erciyes University, three lichenicolous fungi (*Arthonia epicladonia*, *Lichenostigma dimelaenae*, *Sphinctrina leucopoda*) and one lichenized fungus (*Rhizocarpon sublavatum*) are reported from Turkey for the first time. Comments on their habitats, substrata, and key anatomical features are provided for each taxon.

**Key words** — *Ascomycota*, lichens, biodiversity, Trabzon, Yozgat

**Introduction**

In the last 20 years, there have been intensive lichenological studies to determine the lichen mycota of Turkey (e.g. John 1996, Aslan 2000, John & Breuss 2004, Halıcı et al. 2005, Tufan et al. 2005, Candan & Özdemir Türk 2008). At the moment, approximately 1200 lichenized fungal species are known from Turkey but at least 2000 lichenized fungal species are expected in the country (Halıcı et al. 2007a). The checklist of lichenized and lichenicolous fungi of Turkey is being prepared by Volker John and should be published in a few years (V. John, pers. comm.).

The lichenicolous fungi of Turkey have started to receive more attention during the last five years, and a key to the 117 known taxa of lichenicolous *Ascomycota* (including mitosporic fungi) of Turkey was published by Halıcı (2008a). After this publication, there were some more additions (e.g. Candan &

Halıcı 2008, Halıcı 2008b,c, Halıcı & Candan 2009, Halıcı et al. 2009, Candan et al. 2010) and the number of lichenicolous fungal taxa known from Turkey has reached 157. With the 3 species reported in this paper, 160 lichenicolous fungal species are known from Turkey.

### Material and methods

The specimens are deposited in the lichen herbarium of Erciyes University, Biology Department (Kayseri, Turkey). They were examined by standard microscopic techniques. Hand sections were studied in water, potassium hydroxide (KOH) and Lugol's solution (I). Measurements were made in water and the extreme values outside the main range are given in parentheses.

### The species

*Arthonia epicladonia* (Nyl.) Alstrup & Zhurb.

A detailed description is provided by Zhurbenko & Alstrup (2004) and figures were provided by Alstrup & Hawksworth (1990) under the name *Scutula epicladonia* (Nyl.) Zopf.

**TRABZON:** Of, UZUNGÖL-SOĞANLI GEÇIDI, 40°36.117'N, 40°16.682'E, alt. 2110 m, on squamules of *Cladonia pyxidata* on mosses, 30 Sep. 2008, M.G. Halıcı & I. Akata (MGH 0.6320).

*Arthonia epicladonia* was collected on the squamules of *Cladonia pyxidata* from northeast of Turkey. The Turkish specimen seems to be pathogenic as the infected squamules eventually become brownish. Zhurbenko & Alstrup (2004) did not observe any pathogenic effect in the American specimen; they also cited a wider ascospore size range [(10–)14–17.5(–20) × 5–5.5(–6) µm] than we observed in our Turkish specimen (14–15 × (3.5–)4–5 µm). All other Turkish characters agree well with the description given in Zhurbenko & Alstrup (2004).

New to Turkey.

*Lichenostigma dimelaenae* Calat. & Hafellner

A detailed description is provided by Calatayud et al. (2004).

**YOZGAT:** Şefaatli, ŞEKERCİ DAĞI, 39°32.511'N, 34°43.242'E, alt. 880 m, on areoles of *Dimelaena oreina* on siliceous rocks, 12 Jul. 2009, M. Kocakaya (MGH 0.4018).

Ascomata not connected to superficial hyphal strands and forming dense groups, centrum I + pale red. Asci 8-spored, subglobose to globose, 25–28 × 25–28 µm. Ascospores brown, 1-septate, broadly obovate and constricted at the septum, not halonate, 13–16 × 6.5–11 µm.

Previously this species was recorded only from the USA. The Turkish specimen is identical with the original species description. New to Turkey.



***Rhizocarpon sublavatum* Fryday**

A detailed description is provided by Fryday (2000).

**TRABZON:** Of, UZUNGÖL-SOĞANLI GEÇIDI, 40°36.117'N, 40°16.682'E, alt. 2110 m, on exposed siliceous rocks, 30 Sep. 2008, M.G. Halıcı & I. Akata (MGH 0.2920).

The Turkish specimen has a cracked-areolate and brownish-grey thallus, which is clearly limited by a black prothallus. Ascospores are hyaline to very pale brownish, muriform with 19–20 cells, and  $(24- )29-30(-34) \times (11- )13-14 \mu\text{m}$ .

Fryday (2000) noted that *R. sublavatum* has ascospore characters intermediate between *R. reductum* and *R. lavatum* and suggests that it is a northern montane species, probably with some oceanic affinities. The Turkish specimen, which was collected at 2110 m altitude in a very humid locality, supports confirms this observation.

Previously reported only from UK (Fryday 2000) and Norway (Ihlen 2004). New to Turkey.

***Sphinctrina leucopoda* Nyl.**

Detailed descriptions are provided by Löfgren & Tibell (1999) and Tibell (2004).

**YOZGAT:** Akdağmaden, BÜYÜK NALBANT MOUNTAIN, 39°32'N, 36°00'E, alt. 2150 m, on *Lecanora swartzii* on exposed siliceous rocks, 14 Aug. 2004, M.G. Halıcı & M. Kocakaya (MGH 0.4016).

The Turkish specimen is parasymbiotic, has distinctly stalked apothecia, 8-spored asci measuring  $45-53 \times 6-7 \mu\text{m}$ , and non-septate brown ascospores that are minutely ornamented in maturity. Ascospores of the Turkish specimen are slightly larger [ $(5- )5.5-6(-7) \mu\text{m}$  vs.  $(4- )4.3-6.3 \times 4-5.7(-5.8) \mu\text{m}$ ] than the reports previously given for the species (Löfgren & Tibell 1999).

This variable species is sometimes hard to distinguish from *Sphinctrina turbinata* morphologically, but the latter species shows a characteristic K + intensified red pigment in the exciple as stated by Löfgren & Tibell (1999) and Tibell (2004). The Turkish specimen was collected on the areoles of *Lecanora swartzii*, although *S. leucopoda* is also reported frequently on *Pertusaria pertusa* and rarely on *Diploschistes* or *Lecanora* on rocks (Löfgren & Tibell 1999, Tibell 2004). *Sphinctrina leucopoda* is rarely reported on *Lecanora swartzii* from Sweden (Ihlen & Wedin 2008).

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## MYCOTAXON

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**A new species of *Heteroconium* from Fujian, China**

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**Abstract** — *Heteroconium schimae* sp. nov. is described and illustrated occurring on dead branches of *Schima superba*. The specimen was collected from tropical forests in Fujian province of China. The type specimen is deposited in HSAUP (Herbarium of the Department of Plant Pathology, Shandong Agricultural University) and HMAS (Mycological Herbarium, Institute of Microbiology, Chinese Academy of Sciences).

**Key words** — hyphomycetes, taxonomy

**Introduction**

The genus *Heteroconium* was erected by Petrak (1949) with *H. citharexyl* F. Petr as the type species. The generic characteristics of *Heteroconium* include macronematous, mononematous conidiophores which are unbranched or with a few branches originating after conidial secession. The conidiogenous cells are monoblastic, terminal, and proliferate percurrently, and the conidia are dry, euseptate, cylindrical to oblong, sometimes curved, and arise in acropetal unbranched chains (Petrak 1949, Castañeda et al. 1999, Taylor et al. 2001). Conidial secession is schizolytic. These characters also separate the genus from similar genera such as *Lylea* Morgan-Jones, *Xenoheteroconium* Bhat et al., *Cladophialophora* Borelli, *Septonema* Corda, *Phaeoblastophora* Partr. & Morgan-Jones, *Taeniolella* S. Hughes, *Cylindrium* Bonord, and *Hormiactis* Preuss (Castañeda et al. 1999, Kwaśna et al. 2007). To date, 18 taxa have been assigned to the genus *Heteroconium*, although several have been transferred to other genera. *Heteroconium tetracoilum* (Corda) M.B. Ellis (Ellis 1976) was transferred to *Lylea* as *L. tetracoila* (Corda) Hol.-Jech. (Holubová-Jechová 1978), while *Heteroconium solaninum* (Sacc.& P. Syd.) M.B. Ellis (Ellis 1976) was designated as the type species of the genus *Pirozynskiella* S. Hughes (Hughes 2007) based on its obligate association with asterinaceous fungi

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\*Corresponding author

and in the centrifugal sequence of conidium trans-septation after the initial median septum. *Heteroconium chaetospira* (Grove) M.B. Ellis (Ellis 1976) was transferred to *Cladophialophora* as *C. chaetospira* (Grove) Crous & Arzanlou (Crous et al. 2007) following a molecular study of the *Herpotrichiellaceae* and *Venturiaceae*. *Heteroconium queenslandicum* Matsush. (Matsushima 1989) has undifferentiated conidiophores and both mono- and polyblastic conidiogenous cells. It is not congeneric with *Heteroconium* species and is more closely related to the genus *Parapleurotheciopsis* P.M. Kirk (Kirk 1982), although a new combination has not been proposed from China.

The species of *Heteroconium* have been described from a variety of substrates including living or decaying leaves, dead twigs, dead wood, and bark, especially in damp conditions and warmer climates. During a study of tropical microfungi from the forest of Fujian province of southern China, numerous anamorphic fungi were collected. Among them, a previously undescribed species of *Heteroconium* was found which differed in conidial morphology. It is proposed herein as new.

### Taxonomic description

***Heteroconium schimae*** Y.D. Zhang & X.G. Zhang, sp. nov.

FIGURE 1

MYCOBANK MB 518788

*Coloniae in substrato naturali effusae, atro-brunneae. Mycelium partim superficiale, partim immersum, ex hyphis septatis, pallide brunneis, laevibus, 1–2 µm crassis compositum. Conidiophora macronematosa, mononematosa, nonramosa, erecta, cylindrica, recta, laevia, atro-brunnea, 4–10-septata, 59–127 × 4–5.5 µm. Cellulae conidiogenae monoblasticae, terminales, brunnea, laevia, 9–16.5 × 4–5.5 µm. Conidiorum secessio schizolytica. Conidia cylindrica, lata fusiformia usque ad obclavata, frequenter attenuata ad alternus cum terminales, holoblastica, dilute brunneae, laevibus, 0–6-euseptata, 13–44 × 5.5–10 µm. Teleomorphosis ignota.*

HOLOTYPE: on dead branches of *Schima superba* Gardn. & Champ. (*Theaceae*), forest park of Wuyishan, Fujian Province, China. Aug. 16. 2009, Y.D. Zhang, HSAUP H3100 (isotype HMAS 144866).

ETYMOLOGY: in reference to the substrate genus, *Schima*.

Colonies on the natural substratum, effuse, dark brown. Mycelium partly superficial, partly immersed, composed of septate, pale brown, smooth-walled hyphae, 1–2 µm thick. Conidiophores macronematous, mononematous, unbranched, erect, cylindrical, straight, smooth, dark brown, 4–10-septate, 59–127 × 4–5.5 µm. Conidiogenous cells monoblastic, terminal, brown, smooth, 9–16.5 × 4–5.5 µm. Conidial secession schizolytic. Conidia cylindrical, broad fusiform to obclavate, often tapered at one or both the ends, holoblastic, in chains of up to 4, occasionally with a secondary conidium from its neighbors or from conidial secession, pale brown, smooth-walled, 0–6-euseptate, 13–44 × 5.5–10 µm. Teleomorph unknown.

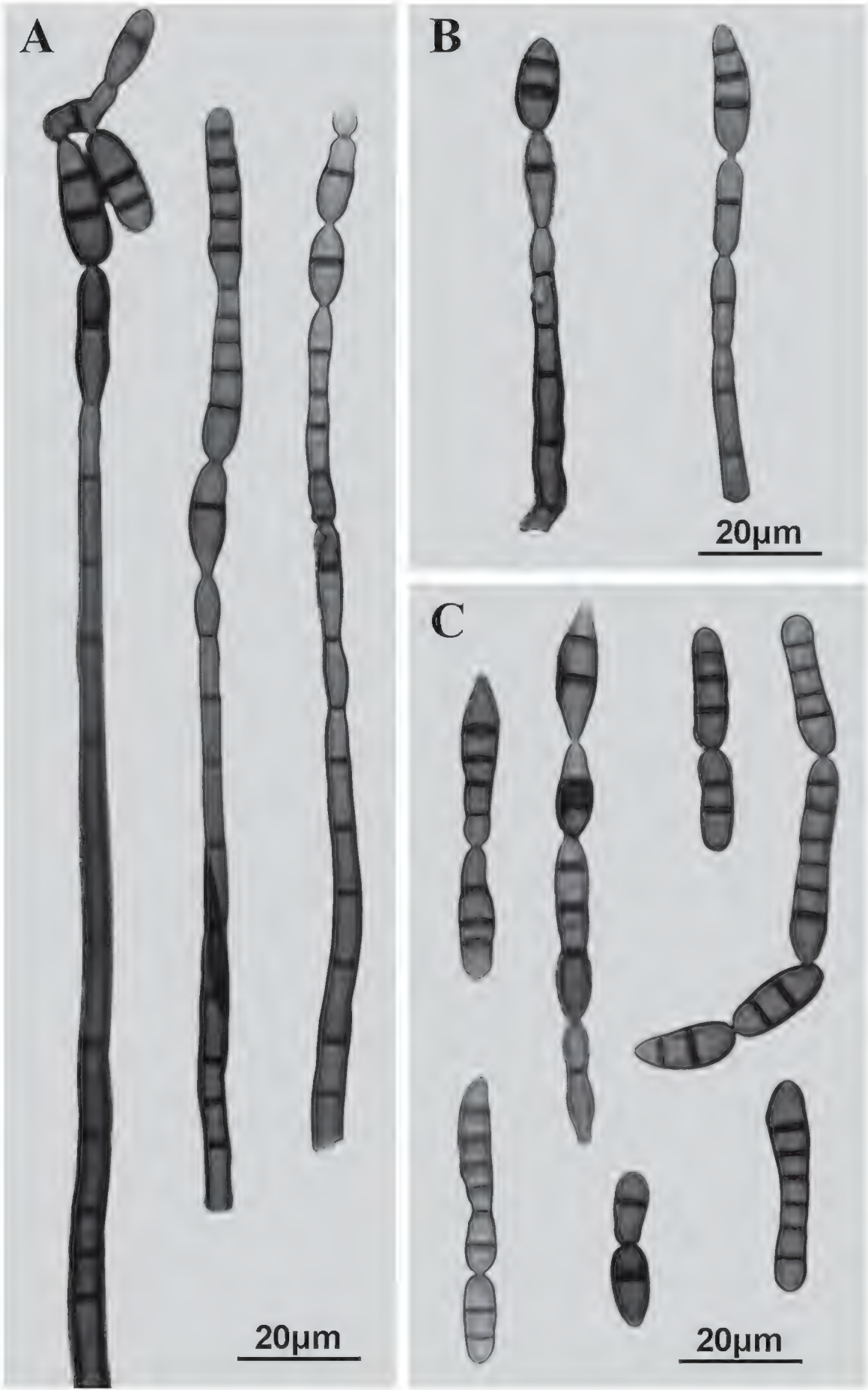


FIG. 1. *Heteroconium schimae*. A–B. Conidiophores with conidia. C. Conidia.



The conidia of *H. schimae* are similar in shape and septation to those of *H. arundicum* Chowdhry (Chowdhry 1980) and *H. citharexyli* (Petrak 1949). However, the conidia of *H. schimae* are smaller than those of *H. arundicum* (35–95 × 8–12 µm), while the conidiogenous cells of *H. citharexyli* are determinate or proliferate percurrently, a feature not found in *H. schimae*. In addition, the conidia of *H. schimae* are in chains of up to 4 and occasionally have a secondary conidium, whereas those are not produced by *H. arundicum* and *H. citharexyli*.

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## MYCOTAXON

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**Development and morphology of *Clathrus delicatus*  
(*Phallomycetidae*, *Phallaceae*) from India**S. SWAPNA<sup>1</sup>, S. ABRAR<sup>1</sup>, C. MANOHARACHARY<sup>2</sup> & M. KRISHNAPPA<sup>1\*</sup>swapnas1007@gmail.com, syedabrar1007@gmail.com  
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**Abstract** — During fieldwork, *Clathrus delicatus* was collected from the Muthodi forest range in the Bhadra Wildlife Sanctuary in the state of Karnataka, India. Although this species was previously recorded from India, these reports did not include detailed morphological descriptions. Here we describe *C. delicatus* and provide illustrations and notes on fruitbody development, which has not been well characterized in the past.

**Key words** — *Phallaceae*, peridial suture, primordia, sporoma, volva-gel

**Introduction**

Members of *Phallales*, commonly called stinkhorns, produce foul-smelling fruitbodies that attract insects. Their distinctive odor is produced by a combination of chemicals such as hydrogen sulfide and methyl mercaptan (List & Freund 1968). Stinkhorns typically develop very quickly, often within few hours, with the spore bearing structures (receptacles) emerging from globose to ovoid structures called ‘myco-eggs’ (Lloyd 1906, Pegler et al. 1995). The order *Phallales* comprises 2 families, 26 genera, and 88 species (Kirk et al. 2008). Clathroid members of family *Phallaceae* form multipileate receptacles (Gäumann 1952) with beautiful and bright colored sporomata. *Clathrus* is unique in having latticed, hollow, spherical or stellate receptacles with slimy glebae (spore masses) borne on their inner surfaces (Pegler et al. 1995). Species in *Clathrus* have simple (Ingold 1971), ellipsoid spores that are typically dispersed after they adhere to the body parts of insects that have been lured to the fruitbody by its fetid aroma (Alexopoulos et al. 2002).

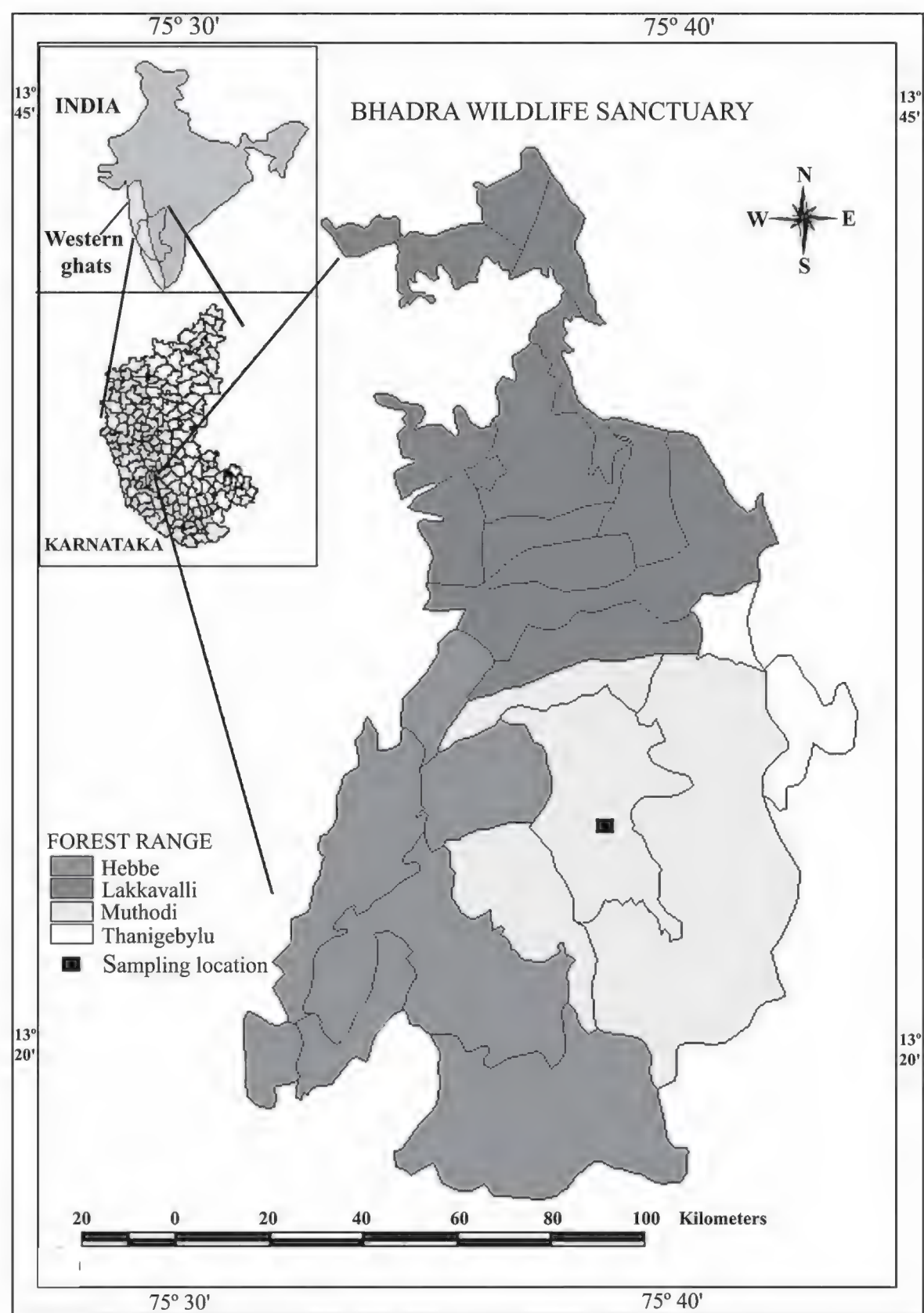


FIG. 1. Sampling location in Muthodi forest range, Bhadra Wildlife Sanctuary.

*Clathrus delicatus* was first described by Berkeley & Broome (1873). Fischer (1898–99) outlined growth stages of *C. delicatus* and compared its receptacle and gleba development to that of *C. chrysomycelinus* Möller. Narasimhan (1932), who published the first report of *C. delicatus* from India (Mysore, Karnataka),

gave a few details on characteristics of the gleba but did not describe the sporoma morphology (e.g., egg and receptacle color and size). Dring (1980) described the development of sporomata in *Clathraceae* (regarded as synonymous with *Phallaceae* by Kirk et al. 2008) and correlated the relationship of receptacle with the other parts of the developing fruitbody. Later, Apte (2005) collected *C. delicatus* during a survey on Owl moths (*Othreis* spp.) in Sanjay Gandhi National Park, Mumbai and sent the photographs to the Smithsonian Institution (USA) for identification but did not provide a morphological description of *C. delicatus*.

The present paper provides the first detailed taxonomic description of *C. delicatus* based on Indian material collected in India, including a systematic study of the sporoma development of this species.

### Materials and methods

Collections were made at the Muthodi forest range in the Bhadra Wildlife Sanctuary, Karnataka, India (FIG. 1), altitude 700 m, temperature 22–28°C and relative humidity 75–90%. Fresh specimens were photographed and color notations were made according to Kornerup and Wanscher (1978). Descriptions of macroscopical characters were compiled from field notes on fresh specimens. Microscopic observations and measurements were made on mounts of receptacle material in 3% KOH stained with 3% phloxine. The primordia were fixed in Pfeiffer's solution containing methanol (absolute) and 40% formalin (w/v) in equal proportions, and then free-hand sections, stained with 1% lactophenol cotton blue and 1% phloxine, were prepared on glass slides for observations under a stereo microscope. The specimens cited are deposited in the herbarium of the Department of Applied Botany, Kuvempu University, Shankaraghatta, Shimoga Dist., Karnataka, India (KUABSAK).

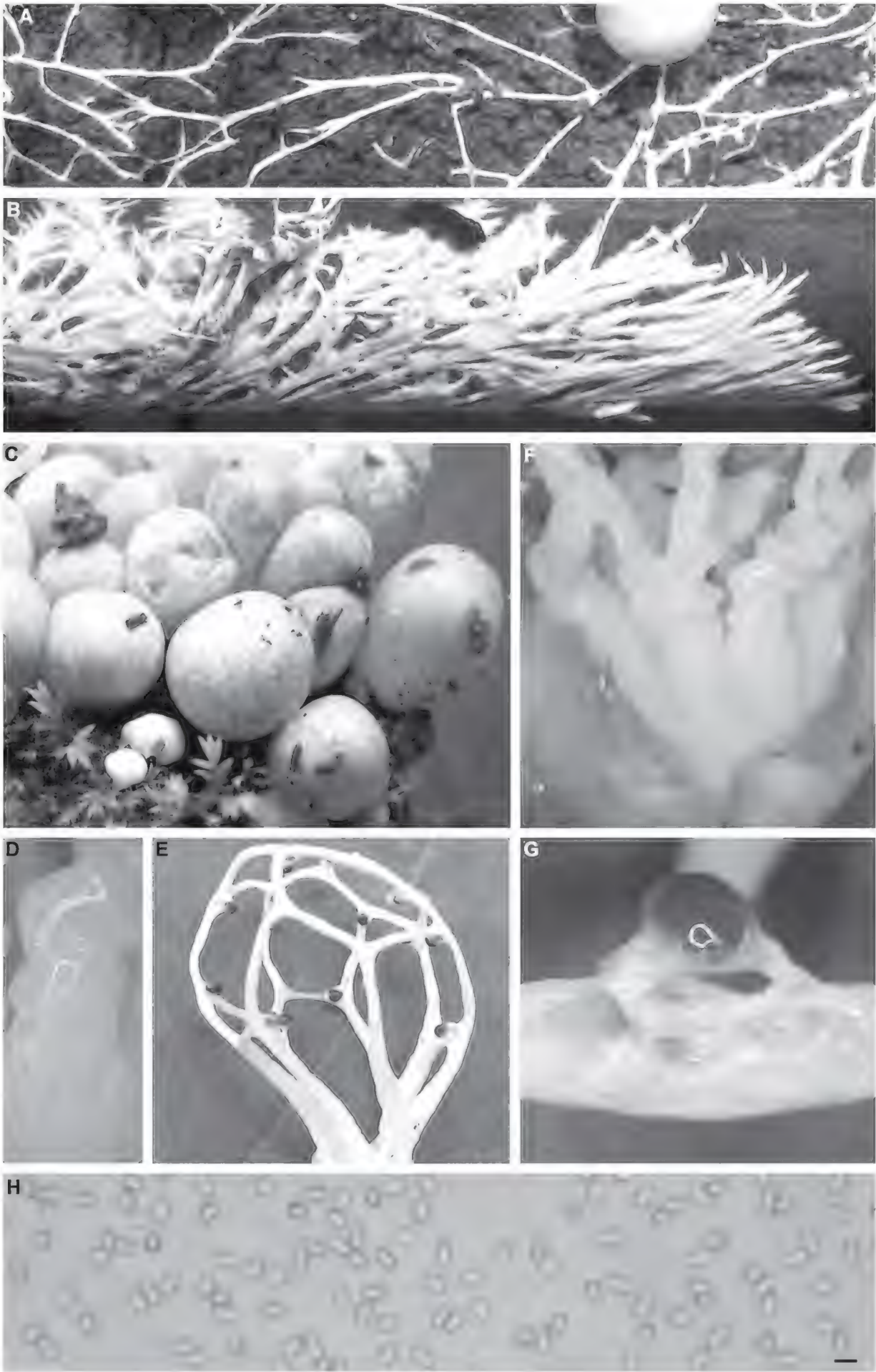
### Taxonomy

*Clathrus delicatus* Berk. & Broome, J. Linn. Soc., Bot. 14: 77, 1873 [“1875”]

FIGS. 2–4

IMMATURE FRUIT BODIES (‘myco-eggs’) arising from thick whitish (1A1) mycelial strands (FIG. 2A) running over twigs (FIG. 2B); globose to ovoid (FIG. 2C), white (1A1) to pale orange (5A1-3), up to 10 mm in diameter, rupturing apically to reveal the expanding receptacle that is initially covered in a mucilaginous substance (FIG. 2D). RECEPTACLE hollow with latticed network, 15–20 × 10–14 mm (FIG. 2E), chalk white (1A1), meshes about 10–12, polygonal, irregularly branched, ± isodiametric towards the apex and vertically elongated towards the base, where arms unite to form a short stipe (FIG. 2F). Arms smooth, flattened, each deeply grooved along their outer-surface. GLEBA







olive brown (4E6), initially coralloid, mucilaginous, deliquescing jelly-like masses restricted to the inner surfaces of the receptacle (toward the apex where arms intersect) on specialized organs (resembling three-legged stools) called glebifers (FIG. 2G). VOLVA pale white to light orange (5A4), thin, enclosing the basal portion of the receptacle. BASIDIOSPORES elliptical,  $1\text{--}2.2 \times 3.6\text{--}4.8 \mu\text{m}$ , smooth, hyaline (FIG. 2H).

SPECIMEN EXAMINED: INDIA, KARNATAKA, Muthodi Forest Range, Bhadra Wildlife Sanctuary (13° 21' 13" N, 75° 38' 10" E, alt. 700m), on decaying vegetation of *Bambusa arundinacea* Retz. (*Poaceae*), 20.VIII.2007, coll. S. Swapna, S. Abrar, C. Manoharachary & M. Krishnappa (KUABSAK-MCH265).

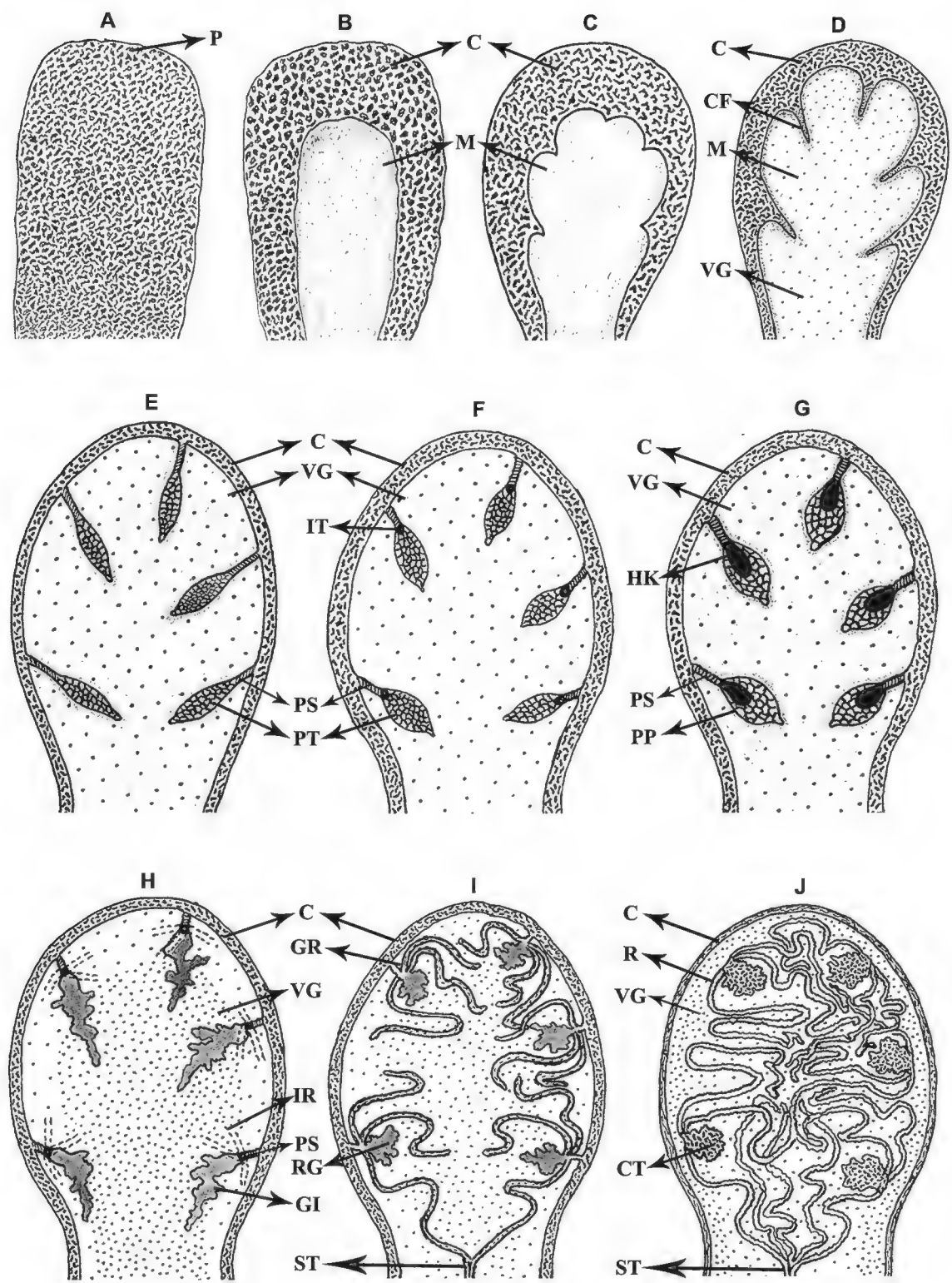
## Development

*C. delicatus* undergoes two phases in the sporomic stage: a myco-egg phase and receptacle phase.

**PRIMORDIA INITIATION:** Primordia initiate at points of swellings along the mycelium strands. The primordium initial (P) lacks an internal structure and is composed of hyphal elements (FIG. 3A). The developing primordium differentiates into central medulla (M) and peripheral cortex (C) (FIG. 3B). The cortical layer develops a series of infoldings (FIG. 3C) that intrude into the inner layer on the medulla. As these infolds become more pronounced, clefts (CF) form and medulla begins to deliquesce, transforming into the volva-gel (VG) within the cortex (FIG. 3D). The primordium increases in size throughout this phase as the cortex and other internal structures develop to form recognizable small myco-eggs.

**MYCO-EGG PHASE:** The clefts further deepen and become compressed, forming peridial sutures (PS) at the myco-egg centres. The deepest point of each peridial suture differentiates into palisade tissue (PT) (FIG. 3E) that comprises the gleba fundamentals. At the peridial suture-palisade tissue junction, an intermediate tissue (IT) develops (FIG. 3F) and then thickens into hyphal knots (HK) while the palisade transforms into pseudoparenchymatous tissue (PP) (FIG. 3G). The hyphal knot begins to divide, branching out on three sides to initiate the receptacle (IR). Each pseudoparenchymatous mass further differentiates to form a glebal initial (GI) (FIG. 3H). The lowermost peridial suture ring producing lower branches proliferates towards the base, each fusing together to form a very short stipe (ST). The growing receptacle (GR) develops further

FIG. 2. *Clathrus delicatus* (KUABSAK-MCH 265). —A White mycelial strands. —B Mycelium covering twigs of *Bambusa arundinacea*. —C A cluster of myco-eggs. —D Mucilaginous substance (volva-gel) coating the emerging receptacle. —E The latticed network of the mature receptacle. —F Arms at the basal portion of the receptacle united to form a short stipe. —G Glebifer. —H Basidiospores. Magnifications: A–C = 15×, D = 40×, E = 12×, F = 35×, G = 45×; scale bar: H = 5  $\mu\text{m}$ .



FIGS. 3A-J. Sporoma development of *Clathrus delicatus* (KUABSAK-MCH 265).

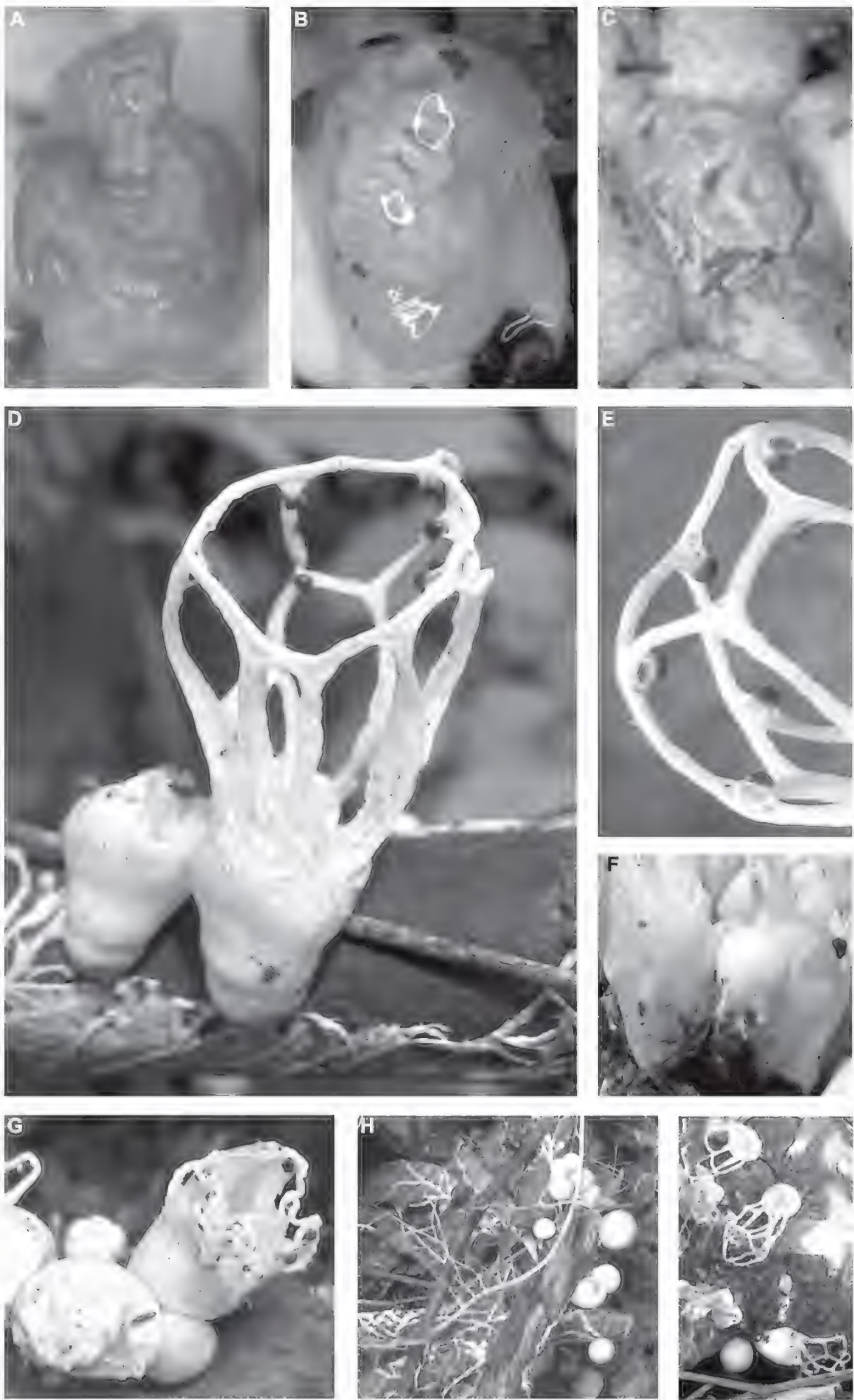
ABBREVIATIONS: C—Cortex, CF—Cleft, CT—Palisade tissue transforming into columella and trama, GR—Growing receptacle, HK—Hyphal knot, GI—Gleba initial, IR—Initiation of receptacle, IT—Intermediate tissue, M—Medulla, P—Primordium, PP—Pseudoparenchyma layer, PS—Peridial suture, PT—Palisade tissue, RG—Reduction of glebal mass, ST—Stipe, VG—Volva-gel.

with the reduction of glebal mass (RG) (FIG. 3I). The continuous development and branching of the receptacle (R) at the centre displaces the volva-gel towards the periphery as the peridial sutures degrade (FIG. 3J) and the central medulla disintegrates. The palisade tissue completely transforms into gelatinized columella and trama (CT), which adheres tightly to the developing receptacle that completely surrounds it (FIG. 4A). After 8–10 days, the volva-gel becomes more viscous (FIG. 4B) as the egg increases in size and basidiospores are formed from hymenial layers forming inverted cup shaped structures (gleba) at the junction of the arms. The mature egg has three distinct layers: the exoperidium (outer skin), mesoperidium (volva-gel), and endoperidium (receptacle and gleba). After the egg ruptures apically (FIG. 4C), the expanding receptacle emerges.

**RECEPTACLE PHASE:** Rupture is caused by increasing turgor pressure and cell elongation in the expanding receptacle. The receptacle freely expands and this phase proceeds rapidly (2–4 minutes) until the mature sporoma has formed (FIG. 4D), with the gleba found at the arm intersections resembling three-legged stools (FIG. 4E). After ‘hatching,’ the ruptured exoperidium remains behind as a volva (FIG. 4F) attached to the mycelial strands. The receptacle eventually shrinks with time (FIG. 4G), and insects attracted by the fetid glebal odor disseminate the spores, thus continuing the life cycle with multiple colonies (FIG. 4H) and developing sporomata (FIG. 4I).

## Discussion

In *Clathrus*, receptacle morphology varies considerably, as does the placement of the gleba within the receptacle. *Clathrus archeri* (Berk.) Dring, *C. crispatus* Thwaites ex E. Fisch., *C. kusanoi* (Kobayasi) Dring, *C. mauritanus* (Lloyd) Dring, and *C. ruber* P. Micheli ex Pers. have gleba distributed over a large portion (with the exception of the more basal areas) of the inner surfaces of the receptacle (Dring 1980, Arora & Burk 1982). In *C. baumii* Henn. and *C. preussii* Henn., the gleba spreads over the inner arm surfaces of the arms but tends to concentrate near where the arms intersect. In *C. columnatus* Bosc the gleba is found only at the more apical portions of the receptacle as a centralized glebal mass that spreads down along the inner surface of the arms (Dring 1980). In *C. chrysomycelinus* and *C. oahuensis* Dring the gleba is restricted to discrete droplets in glebifers seated on the intersection of the arms (Dring et al. 1971, Dring 1980). Finally, although the gleba of *C. delicatus* is also restricted to the arm intersections, the droplets are very minute, and the glebifers are even more specialized in their structure, resembling miniature three-legged stools (Dring 1980).



Dring et al. (1971) suggested that variation found within *Clathrus* could be interpreted in an evolutionary context, which Dring (1980) later placed in several evolutionary “series.” From the “primitive” state, generally these series progressively “simplified” in the distribution of the gleba, accompanied by a reduction in glebal quantity. These trends were also associated with a reduced receptacle size as well as with an increasing complexity in the localization of the gleba, with glebifers occurring in the most advanced forms (Dring 1980). *Clathrus delicatus* was considered one of the more advanced species in *Clathrus*, exhibiting the most specialized and complex glebifer form (Dring et al. 1971, Dring 1980). Here we also document for *C. delicatus* the extremely small receptacle size ( $15\text{--}20 \times 10\text{--}14$  mm), which Dring (1980) also considered a more evolutionarily advanced trait.

A recent molecular phylogeny of the *Phallomycetidae* (Hosaka et al. 2006) included *Clathrus ruber* and *C. chrysomycelinus*, as well as other species in the *Phallales*. Although many early authors (Fischer 1898–99, Lloyd 1906, Petch 1908) suggested that *Clathrus* is the most primitive genus within the *Clathraceae*, Hosaka et al. (2006) placed *Clathrus* species within a more recently derived *Clathraceae* clade that is sister to the *Phallaceae* clade. As Hosaka et al. (2006) only included two species of *Clathrus* in their study, evolutionary relationships among *Clathrus* species remain poorly understood.

### Acknowledgements

The work was supported in part by University Grants Commission (UGC), New Delhi. We are grateful to Dr. D.W. Minter (Principal Scientist at CABI, UK) for very promptly providing us pertinent literature through the online digital library for mycology ([www.cybertruffle.org.uk](http://www.cybertruffle.org.uk)). We thank Drs. P.M. Kirk and P.F. Cannon (Principal Scientists at CABI, UK) for helpful discussions. Furthermore, we greatly appreciate the valuable support of Prof. M.B. Shivanna and Mr. K.G. Somashekhara Achar (Dept. of Applied Botany, Kuvempu University, Shankaraghatta) during the course of this research. We thank Drs. Laura Guzmán Dávalos (Departamento de Botánica y Zoología, Universidad de Guadalajara, México), Vagner G. Cortez (Universidade Federal do Paraná, Brazil), and Scott T. Bates (Fierer Laboratory, University of Colorado, Boulder) for critical review of the manuscript.

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FIG. 4. Sporoma development of *Clathrus delicatus* (KUABSAK-MCH 265). — A Gelatinized tissue adhering to the developing receptacle. — B Volva-gel enveloping the receptacle. — C Apical rupturing of the myco-egg. — D Expanded receptacle. — E Gleba at the intersections of arms. — F Volva. — G An aged receptacle, shrinking with desiccation. — H Mycelium strands with eggs forming intermittently. — I Expanded receptacles of a colony.

Magnifications: A = 60×, B–C, E = 25×, D, F = 20×, G = 10×.



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## MYCOTAXON

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***Catillaria*, *Cladonia*, *Strigula*, and *Cresporhaphis* species  
new to Turkey and Asia**

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**Abstract**—Four species of lichen-forming fungi — *Catillaria atomarioides*, *Cladonia cyathomorpha*, *Strigula brevis*, *Cresporhaphis wienkampii* — are reported as new to Turkey and Asia.

**Key Words**—biodiversity, biota, Giresun

The lichen biota of Turkey is still largely unknown. In the last three years, many new lichen species were reported (e.g. Candan & Özdemir Türk 2008, Çobanoğlu et al. 2008, Halıcı & Aksoy 2009, Kinalioğlu 2009, Öztürk & Güvenç 2010, Yazıcı et al. 2010). This contribution reports four species as first records for Turkey.

Specimens were collected in the provinces of Hatay, Giresun, and Ordu between 17 July 2004 and 10 April 2010. They were identified with various lichen guides (mainly Smith et al. 2009). Vouchers are preserved in the herbarium of the Faculty of Science and Arts, Giresun University, Giresun, Turkey. The collector and collection number are given in parentheses after the locality details.

**Species recorded*****Catillaria atomarioides*** (Müll. Arg.) H. Kiliass

FIG 1

Thallus thin, dark olivaceous to blackish. Apothecia 0.1–0.25 mm diam, black, sparse, mainly plane. Epithecium mostly dark brown to dark green. Hymenium

colourless, 32.5–40 µm tall. Ascospores simple, ellipsoid, colourless, 8–10 × 2.5–3.7 µm. Thallus C–, K–, KC–, PD–.

SPECIMEN EXAMINED: Giresun, Keşap, sea shore, 40°58'20"N, 38°37'23"E, 0 m, 11 Apr. 2010, on siliceous rock (Kinalioğlu 1801).

Known previously from western and northern Europe, Macaronesia, and Africa on hard acid rocks (including river shingle and slate) and brick. In Turkey the specimen was collected from siliceous rock along the coast. New to Asia.

A detailed description is provided by Smith et al. (2009).

DISCUSSION: *Catillaria atomarioides* is easily mistaken for a diminutive form of *C. chalybeia* or *C. subviridis* (coastal, pale inner roper margin), or even *Amandinea punctata* (Smith et al. 2009). The hymenium is slightly smaller in the Turkish specimen than in the European, Macaronesian, and South American material. Original descriptions of this species report hymenium up to 30–40 µm (Smith et al. 2009). The Turkish collection differs ecologically by occurring only at coastal localities.

*Cladonia cyathomorpha* Stirt. ex Walt. Watson

FIG 2

Primary thallus dominant. Squamules 2–4 mm broad, greenish above, white below. Podetia rare, up to 2–5 mm tall, forming cups to 3 mm wide, coarsely corticate within. Thallus C–, K+ yellow, KC–, PD+ red.

SPECIMENS EXAMINED: Giresun, Keşap, 40°58'22"N, 38°37'36"E, 4 m, 12 Feb. 2006, on siliceous rock (Kinalioğlu 1804). Ordu, N of Ünye, Çamlık, sea shore, 2 m, 21 Jul. 2006, on soil (Kinalioğlu 1805).

Known from western Europe, Macaronesia, and South America, mostly on vertical faces of mossy rocks in hilly and montane areas. New to Turkey and Asia. In Turkey the specimens were only collected from siliceous rock and soil along the coast.

A detailed description is provided by Smith et al. (2009).

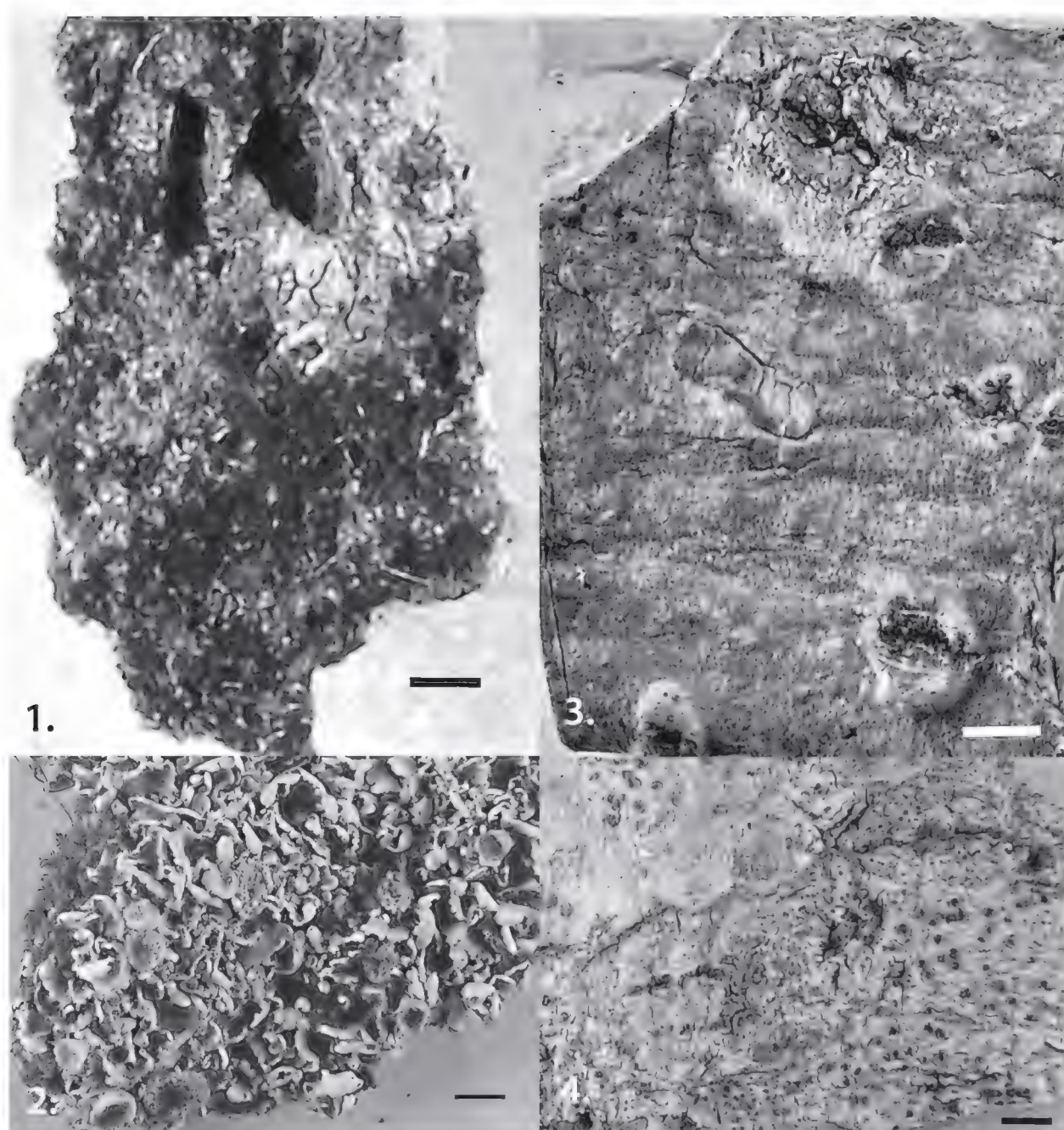
DISCUSSION: *Cladonia cyathomorpha* is distinguished from *C. pyxidata* in having larger, veined, basal squamules and an additional unidentified compound with fumarprotocetraric acid (Smith et al. (2009). The Turkish material is distinguished from western European, Macaronesian, and South American specimens by smaller podetia and squamules. Smith et al. (2009) cite podetia as to 0.8 cm, basal squamules 5–10 mm diam. The Turkish collection differs ecologically in occurring both on siliceous rock and on soil at coastal localities.

*Cresporhaphis wienkampii* (J. Lahm ex Hazsl.) M.B. Aguirre

FIG 3

Thallus embedded in bark cells. Perithecia black, superficial, to 0.2–0.4 mm diam. Ascospores 22.5–30 × 3–3.5 µm in size, colourless. Thallus C–, K–, KC–, PD–.





FIGS. 1–4. Habitus of four lichens new to Turkey. *Catillaria atomarioides* (Kınalıoğlu 1801). FIG. 2. *Cladonia cyathomorpha* (Kınalıoğlu 1804). FIG. 3. *Cresporhaphis wienkampii* (Kınalıoğlu 1814). FIG. 4. *Strigula brevis*, (Kınalıoğlu 1811). Scales: 2 mm.

SPECIMEN EXAMINED: **Hatay**, Dört Yol, S of Konak Village, 36°48'29"N, 36°15'10"E, 172 m, 01 Feb. 2008, on *Quercus* sp. (Kınalıoğlu 1814).

Previously known only from Europe. On living bark. New to Turkey and Asia.

A detailed description is provided by Smith et al. (2009).

DISCUSSION: The perithecia in the Turkish collection are slightly larger than in the European specimen, where the perithecia measure 0.15–0.3 mm diam.

*Strigula brevis* Bricaud & Cl. Roux

FIG 4

Thallus grey-white, partly immersed. Perithecia black, hemispherical, almost semi-immersed, 0.2–0.5 mm diam. Ascospores  $25\text{--}37.5 \times 5\text{--}7.5 \mu\text{m}$ , 3–5 septate, fusiform. Thallus C–, K–, KC–, PD–.

SPECIMEN EXAMINED: Ordu, Gülyalı, Turnasuyu village, 41°03'20"N, 37°59'04"E, 17 m, 17 Jul. 2004, on *Juglans regia* (Kinalioğlu 1811).

Known from western Europe and Macaronesia (Roux & Sérusiaux 2004). On living bark. New to Turkey and Asia.

A detailed description is provided by Roux & Sérusiaux (2004).

DISCUSSION: The perithecia and ascospores of the Turkish material are larger than in the western European and Macaronesian collections, where perithecia are 0.2–0.3 mm wide and ascospores measure  $(17)18\text{--}23.5(25) \times 3.5\text{--}4.5 \mu\text{m}$ .

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***Lactarius fumosibrunneus* in a relict  
*Fagus grandifolia* var. *mexicana* population  
in a Mexican montane cloud forest**

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**Abstract** — *Lactarius fumosibrunneus*, a species considered in the literature congeneric with *L. fumosus*, is interpreted here as an independent taxon due to the differences in the structure of pileipellis and presence of cystidia. Recognition of *L. fumosibrunneus* is supported by morphological comparison with original collections, Mexican samples, and type specimens of related taxa. Collections of *L. fumosibrunneus* were found in the Mexican montane cloud forest of Central Veracruz (east coast of Mexico) where it appears to be ectomycorrhizal partner of the tree *Fagus grandifolia* var. *mexicana*.

**Key words** — ectomycorrhizal fungi, *Fagaceae*, neotropical fungi, *Russulaceae*, taxonomy

## Introduction

*Lactarius fumosibrunneus* A.H. Sm. & Hesler is an American member of subgenus *Plinthogalus* (Burl.) Hesler & A.H. Sm. described by Smith & Hesler (1962) from Michigan, U.S.A. Based on the macroscopical resemblance of *L. fumosibrunneus* with *L. fumosus* Peck, Hesler & Smith (1979) considered it as conspecific. During a regular monitoring of the Mexican montane cloud forest in Veracruz (east coast of Mexico) by the authors (Montoya et al. 2010), some populations of a taxon macroscopically close to the aforementioned species were observed. After a comparative study of collections of these populations with specimens from U.S.A. (including type materials) of *L. fumosibrunneus*, *L. fumosus*, and *L. fumosoides* A.H. Sm. & Hesler, we found that based on differences in the nature of the pileipellis and cheilocystidia, *L. fumosibrunneus* appears distinct from other allied taxa. We therefore consider *L. fumosibrunneus* to represent an independent taxon and support the original concept as published by Smith & Hesler (1962).

## Materials & methods

Monitoring was conducted between September 2006–09 in Acatlán Volcano, Central Veracruz (east coast of Mexico). Samples of *Lactarius* were gathered during random field trips in a stand of *Fagus grandifolia* var. *mexicana*. Collections are kept in XAL herbarium. Basidiomes were studied in fresh condition. Colors were compared with those from Kornerup & Wanscher (1967), e.g. codified as 5D5–E5, and Munsell color chart (1994), e.g. 10YR 4/3–4/4. For the study of micromorphological features, hand sections of dried specimens were rehydrated in 3% KOH. Basidiospores (measurement, shape and ornamentation pattern) were observed in Melzer's reagent. Methods to determine spore ranges are those used by Montoya & Bandala (2003). In the basidiospore descriptions, Xm indicates the range of means of basidiospore length and width and Qm indicates the range of the means of Q (length/width ratio) from n collections (25–50 basidiospores were measured per collection then X indicates their mean). Line drawings were made with the aid of a drawing tube. Acronyms for herbaria follow Holmgren & Holmgren (1998).

## Taxonomy

*Lactarius fumosibrunneus* A.H. Sm. & Hesler, Brittonia 14: 439, 1962      FIGS 1–4A

SPECIMENS EXAMINED. MEXICO. VERACRUZ: Acatlán, ACATLÁN VOLCANO, 14 Sep 2006, Montoya 4625, Montoya 4631, 4633, 4634, 4635; 18 Sep 2007, Montoya 4669; 19 June 2008, Montoya 4680; 30 July 2009, Montoya 4739, Montoya 4740, Montoya 4745 (all at XAL).

PILEUS 12–65 mm diam., convex, becoming plane to plano-convex, depressed in the center with age, at times subumbonate, with or without a central papilla, faintly velutinous, dull, smooth when young to rugose at center when mature or at times venose-rugose and faintly rugose in other areas, dry, firm, often pale greyish (10YR 5/4–6/4, 10YR 5/3) or brownish (5D5–E5) or with darker (5E5–E4–E6, 10YR 4/3–4/4) shades but generally conserving paler or even cream colored patches or appearing with greyish-brown tinges over a cream background or more or less uniformly greyish-brown or brownish (around 2.5Y 5/3–4; 6F7, 5B3–C4; pale 5D5–E5, 4B2), darker (7.5YR 4/3, 5E4–E5) towards the center; margin wavy, at times inflexed and irregular, lobulate, edge at times whitish. LAMELLAE narrow (2–3 mm broad), crowded, short-decurrent to decurrent, cream-colored (2.5Y 8/2–3, 3–4A2) when young to yellowish-ochraceous (5A3, 10 YR 8/3–4, 8/6, 7/6) when old, staining reddish-salmon (8A5–B7) when cut, some furcate, with lamellulae of different length (frequently one longer and two very short), generally 1–3 between two lamellae. STIPE 20–75 × 3–12 mm, subcylindrical, slender, more or less tapering downwards or with tapered base, almost straight, at times weakly sinuous, occasionally curved, firm, hollow



FIG. 1. *Lactarius fumosibrunneus* Montoya 4669. Scale bar= 10 mm.

with age, faintly velutinous, dry, dull, whitish, bone-whitish to cream-colored (2.5 Y 7/3, 8/2, 3A2), later developing pale greyish-brown (5B3–C3–D4, 10YR 5/4–6/4) shades but conserving whitish areas mainly at apex or base; base whitish. CONTEXT white to cream-colored, staining pinkish, becoming slowly reddish (9B3, 9C5), finally wine-red to salmon color (7B5–6, 7A5). ODOR mild to somewhat similar to chlorine. Taste very hot. KOH negative on pileus and context. LATEX white, unchanging, cut surfaces staining reddish, salmon-red (7A6–B6) or even brownish-red (9C8), dried drops stained reddish, staining white paper red (8C5–6), spots on paper slowly turning orange to salmon color (8B5) and to yellow (4A8–A5) with reddish-orange tinges to totally yellow (4A2–3) after some hours.

BASIDIOSPORES  $7-8(-8.5) \times 6.5-7.5(-8) \mu\text{m}$ ,  $X_m = 7.4-7.6 \times 6.5-7.2 \mu\text{m}$ ,  $Q_m = 1.06-1.07$ , subglobose, ornamentation  $1-2 \mu\text{m}$  high, subreticulate, composed of broad, sinuous bands forming a somewhat wide mesh, more or less crestate in profile, at times with isolate verrucae, often weakly amyloid in the suprahilar area. BASIDIA  $42-58 \times 9-13 \mu\text{m}$ , clavate, bi- or tetrasporic, sterigma  $4-7 \mu\text{m}$  long. CHEILOCYSTIDIA  $19-50 \times 5-7.5 \mu\text{m}$ , subcylindrical, more or less narrowly lageniform or moderately tapered, apically rounded,

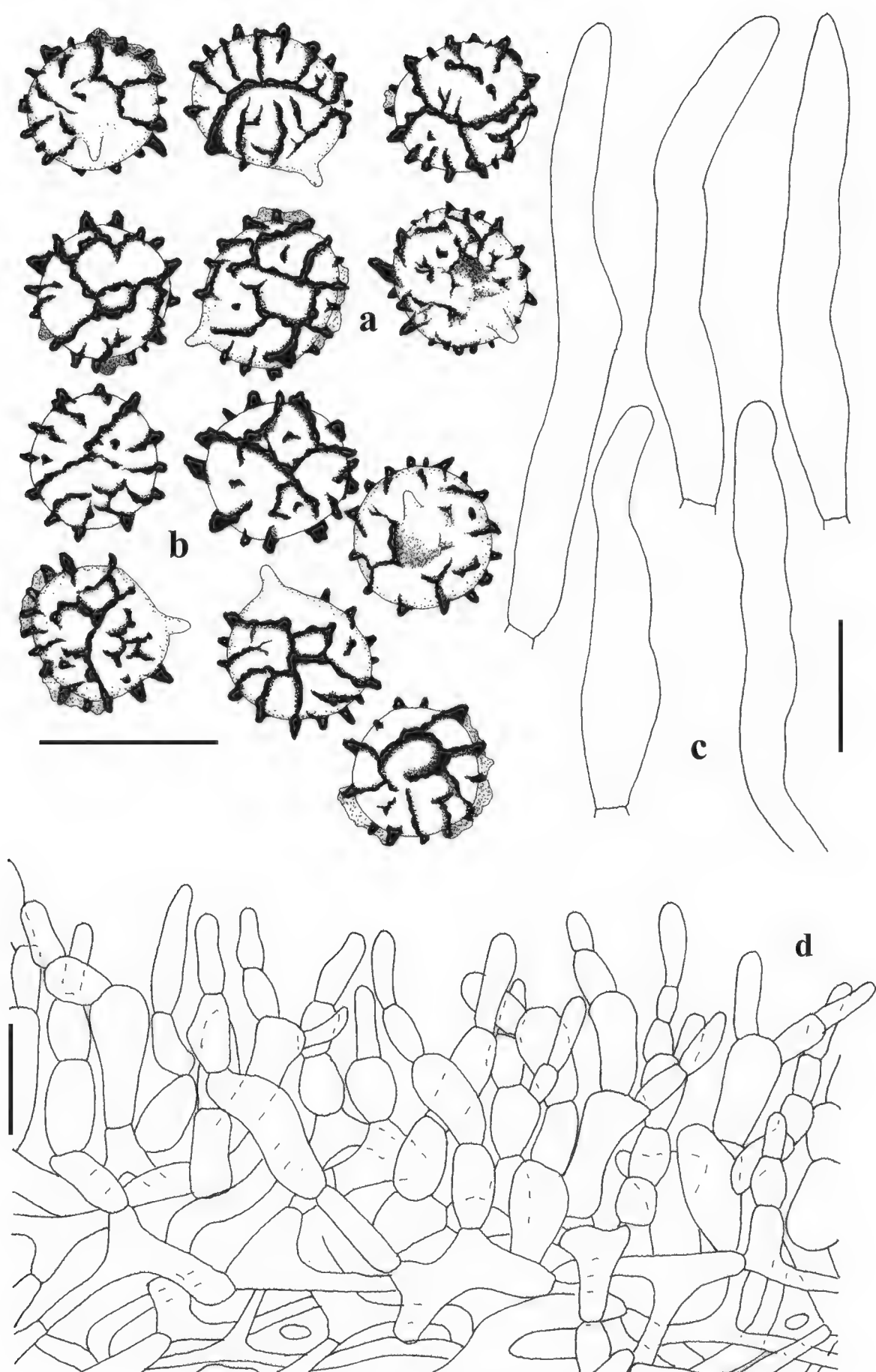


FIG. 2. *Lactarius fumosibrunneus*.  
(a–b) basidiospores, (c) cheilocystidia, (d) pileipellis.  
[a,c,d= Montoya 4634, b = holotype.] Bars: a–c = 10  $\mu$ m, d = 20  $\mu$ m.



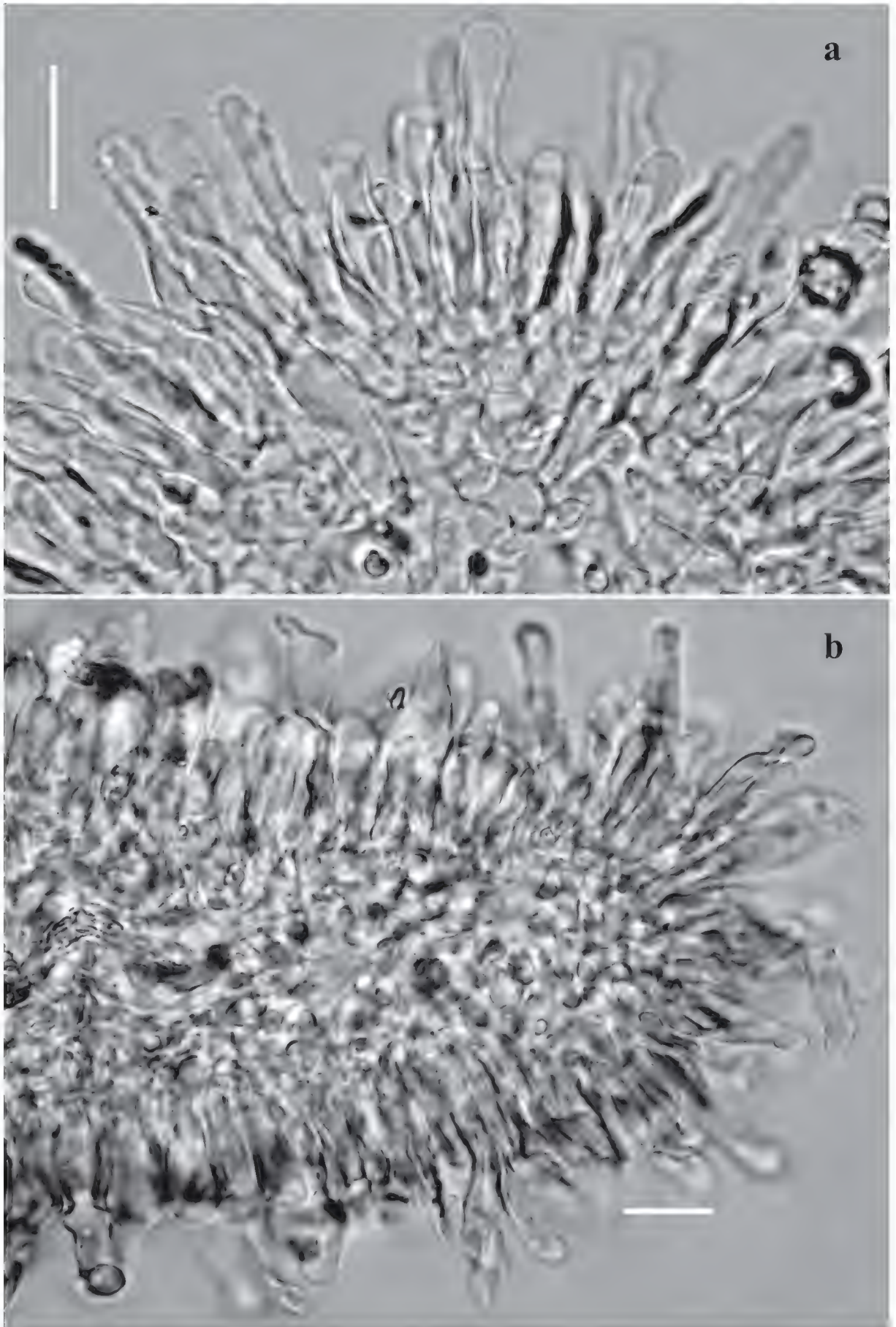


FIG. 3. *Lactarius fumosibrunneus*. Lamellar margin, Montoya 4634. Bars = 20  $\mu$ m.



sinuous, abundant, emerging above hymenium level, hyaline. PLEUROCYSTIDIA absent. PSEUDOCYSTIDIA 3–4  $\mu\text{m}$  diam., subcylindrical to vermiform, at times ramified, with refringent colorless contents. PILEIPELLIS a hymenoeppithelium, 40–62  $\mu\text{m}$  broad, the elements disposed in anticline chains of 2–4 elements long, cells with pale yellowish–brown contents; terminal cells 11–27  $\times$  5–7  $\mu\text{m}$ , subcylindrical, subventricose, pyriform, sinuous, the remaining cells in the chains versiform, those immediately below the terminal element in general broadly subcylindrical, 8–10  $\times$  5–7  $\mu\text{m}$ , other subisodiametric 9–15  $\mu\text{m}$  diam. or more or less versiform and broad, 10–25  $\times$  8–13  $\mu\text{m}$  diam. CONTEXT heteromerous, hyphae 2.5–10.8  $\mu\text{m}$  diam., sphaerocytes 18–39.6  $\mu\text{m}$  diam., laticifers 2.5–11  $\mu\text{m}$  diam. HYMENOPHORAL TRAMA heteromerous, hyphae 3–5  $\mu\text{m}$  diam, laticifers 2.5–6  $\mu\text{m}$  diam., with a lax tissue towards lamellar edge.

HABITAT — Gregarious in a *Fagus grandifolia* var. *mexicana* forest at 1840 m.

OTHER SPECIMENS EXAMINED. USA. MICHIGAN: Washtenaw Co., SHARON HOLLOW, 13 Aug 1960, A.H. Smith 62897 (as *L. fumosus*, MICH); Cheboygan Co., Reese's Bog, 27 Aug 1960, A.H. Smith 63040 (holotype of *L. fumosoides* MICH); Cheboygan Co., Colonial Point, Burt Lake, 11 Aug 1961, A.H. Smith 63892 (holotype of *L. fumosibrunneus*, MICH). NEW YORK, Sandlake, Rensselaer Co., July, Peck s.n. (as "*L. fuliginosus* var. *fumosus* Peck", NYS).

## Discussion

After comparing Mexican materials with specimens and descriptions of *L. fumosibrunneus*, *L. fumosus*, and *L. fumosoides*, we concluded that although they are apparently phenotypically similar, these three taxa could be differentiated because each possesses a unique set of characters. *Lactarius fumosibrunneus* as observed in the type specimen (Smith 63892) presents abundant cheilocystidia distributed at lamellar edges and even placed towards lateral sides of the lamellar margin (Smith & Hesler 1962: '...abundant and extending a short distance up the sides...'); their size and shape (20–57  $\times$  5–7.5  $\mu\text{m}$ , ventricose, subcylindrical, clavate, sinuous) are also similar to Mexican collections (FIGS. 2C, 3A–B). Its basidiospores are 7.3–8  $\times$  6.5–7.5  $\mu\text{m}$ ,  $X = 7.6 \times 7 \mu\text{m}$ ,  $Q = 1.1$ , subreticulate (FIG. 2B). Although the pileipellis (FIG. 4B) was somewhat difficult to rehydrate in the type, it was possible to observe that, as in our Mexican specimens, it is built of groups of elements in chains, basal cells appearing irregular and subisodiametric and the terminal elements having a hymeniform appearance (FIG. 4A). The taste (described as 'burning acrid' by Smith & Hesler 1962) and narrow and crowded lamellae (also observed in Mexican collections) are distinctive. Smith & Hesler (1962) recorded *L. fumosibrunneus* from a beech–maple forest in Michigan.

According to the description by Peck (1872), *Lactarius fumosus* possesses a pileus that is convex and then expanded, slightly depressed in the center,

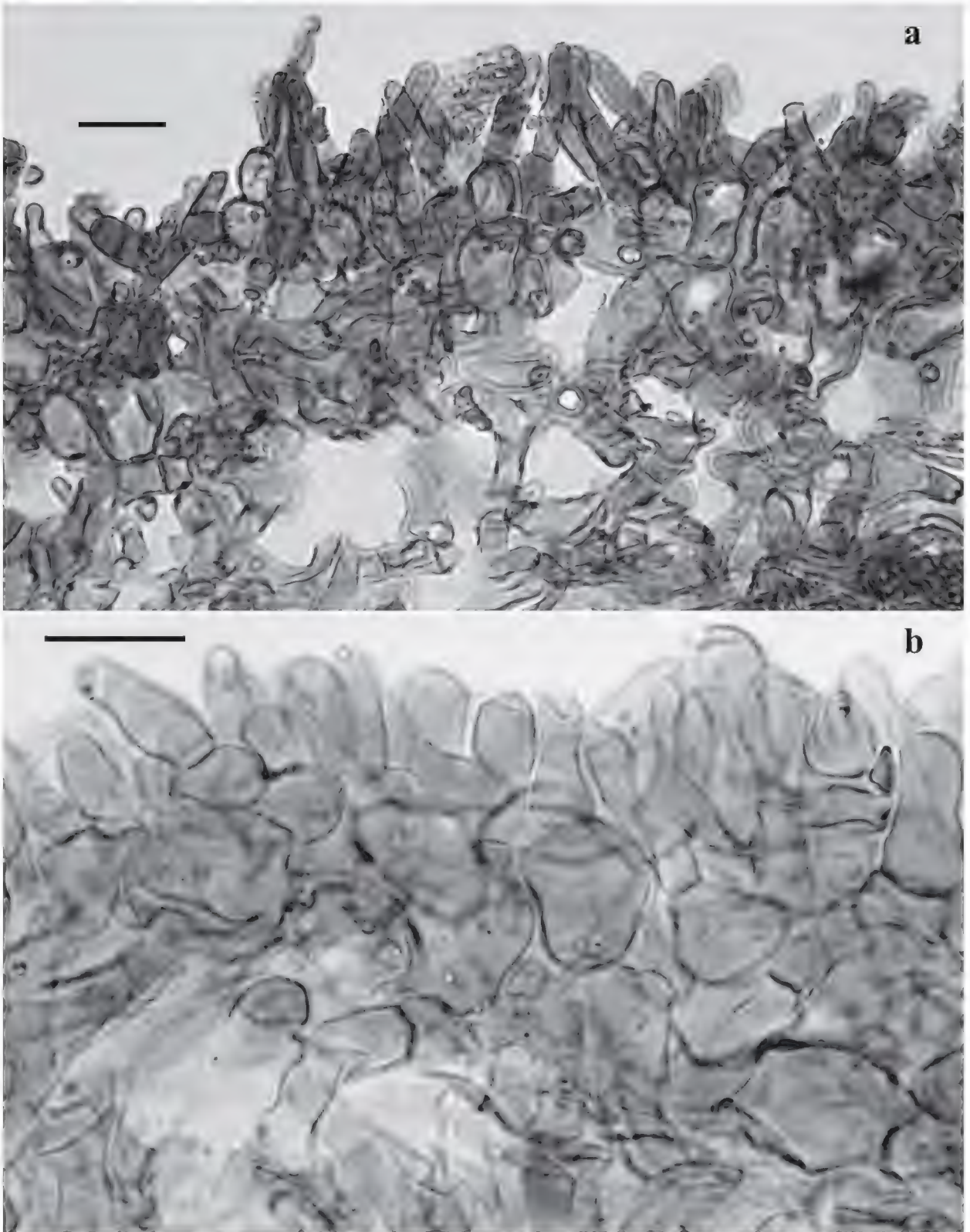


FIG. 4. Pileipellis. Bars = 20  $\mu$ m.  
(a) *Lactarius fumosibrunneus*, Montoya 4634, (b) *L. fumosus*, Smith 62897.

smooth, smoky-brown or sordid white, lamellae close, adnate, flesh white, taste at first mild then acrid. Smith & Hesler (1962) distinguished *L. fumosus* from *L. fumosibrunneus* because basidiomes of the latter are quickly burning-

acid and have a more highly developed pileipellis structure (also observed for the Mexican collections). Subsequently however, Hesler & Smith (1979) synonymized their *L. fumosibrunneus* with *L. fumosus*, regarding the taste and characteristics of the pileipellis (and stipitipellis) as "... slight quantitative variations..." for recognizing two taxa. They also noted that the cheilocystidia in *L. fumosus* were 'poorly differentiated' [(9–)26–36 × 4.5–6 µm]. It has not been possible to study the type of *L. fumosus*, which according to NYS is apparently lost. For the taxonomic interpretation of *L. fumosus* we examined Peck's specimen (July, NY, Sandlake, Rensselaer Co.; see below) that he identified as *L. fuliginosus* var. *fumosus* and Smith 62897, which Hesler & Smith (1979) considered conspecific with *L. fumosus*. We corroborated in both materials that the lamellar edges lack cheilocystidia and, indeed, bear some basidia and sterile basidiole-like cells (FIGS. 5A–B) (the longest about 10–25 × 3.5–9 µm in the specimen of Peck from Sandlake and 17.5–32.5 × 5–8 µm in the specimen Smith 62897) that could not be considered differentiated cells representing cystidia. The pileipellis (FIG 4B) showed the differences as well, having broader and shorter terminal elements [12–21(–28) × 5–12 µm, broadly clavate, ovoid, subisodiametric and less frequently pyriform]. The basidiospores appear more ellipsoid in both Peck's Sandlake specimen [7–8 (–8.5) × 6.5–7.5(–8) µm,  $X = 7.7 \times 6.7$  µm,  $Q = 1.2$ ,  $n = 25$ ] and Smith 62897 [7–5–8 × 6.5–7.3 µm,  $X = 7.8 \times 6.8$  µm,  $Q = 1.15$ ,  $n = 25$ ].

The type specimen of *Lactarius fumosoides* (treated as *L. fumosus* var. *fumosoides* by Hesler & Smith 1979) was also studied for comparison. This specimen differs from the previous specimens particularly in pileipellis structure and the absence of cheilocystidia. The lamellar edges bear basidiole-like structures and some basidia but no differentiated cystidia. The pileipellis has a lax arrangement, which in some areas appears as a cutis from which some slender pileocystidia [19–68 × 5–7 µm, clavate, subcylindrical-vermiform, sinuous, capitate, these latter 9–10 µm broad at apex] appear intermixed. In most areas the pileocystidia grow from irregular (17–68 × 8–15 µm) or somewhat subisodiametric (15–20 × 15–18 µm) elements arranged in chains of up to two cells. The pileocystidia in *L. fumosoides* (type specimen) are long and slender and somewhat resemble a trichodermis and thus differing from those seen in the other collections of *L. fumosibrunneus*.

We therefore agree with Smith & Hesler (1962) that *L. fumosibrunneus* represents a distinct taxon based on the pileipellis structure, consistent presence and shape of cheilocystidia, the size, shape, and ornamentation of basidiospores, color changes and taste of basidiomes, and the shape and disposition of lamellae. It should be noted that the hot taste seems to be directly associated with latex in that basidiomes lacking latex tasted mild or at least less acid than the basidiomes with latex.



It is interesting to note that after Peck (1885) treated *L. fumosus* as *L. fuliginosus* (Fr.) Fr., Saccardo (1887) reduced Peck's taxon to a variety, as "*L. fuliginosus* var. *fumosus* Peck". The European *Lactarius fuliginosus* (Fr.) Fr. and *L. azonites* (Bull.) Fr. (another species within this group), which share a more or less similar habit with *L. fumosibrunneus*, can be distinguished by moderately distant gills, mild or bitter to slightly acrid latex (Heilmann-Clausen et al. 1998, Basso 1999), bigger basidiospores [ $X = 8.0\text{--}8.6 \times 7.4\text{--}7.8 \mu\text{m}$  (in *L. azonites*) or  $X = 8.1\text{--}8.4 \times 7.1\text{--}7.6 \mu\text{m}$  (in *L. fuliginosus* with a wider Q range, 1.09–1.15; Heilmann-Clausen et al. 1998], and a pileipellis with somewhat longer terminal elements that give a trichodermoid aspect to the suprapellis ( $20\text{--}40 \times 3\text{--}5 \mu\text{m}$  in *L. azonites* and  $20\text{--}45 \times 5\text{--}8 \mu\text{m}$  in *L. fuliginosus*; Heilmann-Clausen et al. 1998).

### Acknowledgments

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## MYCOTAXON

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***Hygrocybe manadukaensis* sp. nov. in section *Firmae*  
from Western Ghats, India**

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**Abstract** — A new species *Hygrocybe manadukaensis*, in section *Firmae* collected from the Uppangala forest of Western Ghats of Karnataka, India is described and illustrated. Both macro- and microscopical features of the present collection are compared with similar or closely related taxa in section *Firmae*.

**Key words** — *Agaricales*, *Basidiomycota*, *Hygrophoraceae*, macrofungi

## Introduction

Members of *Hygrocybe* (Fr.) P. Kumm. with dimorphic basidiospores and basidia in section *Firmae* are widely distributed in tropics. Corner (1936) studied this group in the paleotropics and described a new species, *Hygrophorus hypohaemactus* Corner [= *Hygrocybe hypohaemacta* (Corner) Pegler], and 16 new varieties of *Hygrophorus firmus* Berk. & Broome [= *Hygrocybe firma* (Berk. & Broome) Singer]. He also noted, however, that many of his varieties might represent species in their own right. Pegler (1983) stated that *Hygrocybe firma* represented an extremely variable species; he considered that of Corner's varieties, only *militaris* and *puniceoides* (in addition to the autonymous variety) were worthy of recognition at the species level, but he did not transfer any varieties to *Hygrocybe*. Although *Hygrocybe* species are well represented all over India (Manjula 1983, Natarajan et al. 2005), most have been described and reported from Kerala state (Leelavathy et al. 2006). However, only two species of *Hygrocybe* in section *Firmae* have been described so far from India:

*H. alwisii* (Berk. & Broome) Pegler from Kerala (Leelavathy et al. 2006), and *H. natarajanii* Senthil. & Kumaresan from Karnataka (Senthilarasu et al. 2010). In this study, we describe *Hygrocybe manadukaensis*, which differs macro- and microscopically from known species of *Hygrocybe* in section *Firmae*.

## Materials and methods

The description and illustrations were based on the type specimen collected from Manaduka, Uppangala forest of Western Ghats of Karnataka. Handmade sections were obtained from the dried specimens, later revived in 3% KOH and mounted in 2% Phloxine. Approximately 50 basidiospores obtained from a spore print were measured. The mean spore measurements are given in parentheses followed by the range of spore measurements (with extreme values in parentheses). The type specimen was deposited in the Herbarium of Madras University Botany Laboratory (MUBL). The colour terminology used is that of Kornerup & Wanscher (1978).

## Taxonomy

*Hygrocybe manadukaensis* Senthil., Kumaresan & S.K. Singh **sp. nov.**      FIGS 1, 2

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*Pileus* 8–25 mm diam., convexus, depressus; superficie aequabiliter aurantiacus cum flavus tinctus ad discum primus, aurantiacus ad discum, aurantiacus-rubus alibi, flavus ad margine, laevis; margine regularis, laevis, non-striatus. *Lamellae* subdecurrentes, luteolus ad ranunculinus, ad usque 3 mm latae, subdistantes, tribus ordinibus lamellarum intermixtae; margine concolori, laevis. *Stipes* 13–60 × 7–12 mm, aequalis, cylindricus, compressus ad apicem, cavus, caespitosus; superficie aequabiliter aurantiacus-rubus ad atroaurantiacus, laevis. *Contextus* ad usque 2 mm latae at discum, albus. *Sporae* dimorphae; macrospora (12.8 ± 0.7 × 7.8 ± 0.7), (11–)11.5–13.5(–15) × 7–9(–10) µm, Q = 1.6, ellipsoideae ad late ellipsoideae, hyalinae, parietibus tenuibus, guttulis refractives; microspora (5.5 ± 0.4 × 3.4 ± 0.2), (4.5–)5–6(–6.5) × 2.9–4 µm, Q = 1.6, ellipsoideae ad late ellipsoideae, similis ad macrospora. *Basidia* dimorpha; macrobasidia 42.5–57 × 10–13 µm, cylindrico-clavata, 4-spore, sterigmatus 5.5–9.5 × 1.5–2.5 µm, parietibus tenuibus, guttulis numerosis; microbasidia 29–39 × 5.5–6.5 µm, cylindrico-clavata, 4-spore, sterigmatus ad usque 5.5 µm longus, similis ad macrobasidia. *Margo lamellaris* fertilis. *Cystidia* nulla. *Trama hymenophoralis* regularis, ex hyphis 1.5–7.5 µm diam. *Pileal contextus* ex hyphis 1.5–5 µm diam., hyalinae, parietibus tenuibus. *Pileipellis* cutis est ex hyphis repentibus, 1.5–7.5 µm diam. *Fibulis* abundantibus.

**TYPE:** India, Karnataka State, Manaduka, Uppangala Forest, 12°30'N 79°39'W, 500 masl, on ground (soil), Senthilarasu G. (**Holotype** MUBL 3429).

**ETYMOLOGY:** This species is named for its place of collection.

*Pileus* 8–25 mm diam., broadly convex, soon depressed at the disc; surface uniformly deep orange (6A8), with light yellow (4A5) tints at the disc when young, light orange (5A5) at the disc, orange-red (8B8) elsewhere, deep yellow



FIG 1. *Hygrocybe manadukaensis*.  
Under natural conditions in Manaduka, Uppangala forest. Photo Senthilarasu G.

(4A8) at extreme margin with age, dry, smooth; margin regular, smooth, not striate. Lamellae subdecurrent, light yellow (4A4) to butter-yellow (4A5), up to 3 mm broad, moderately close with lamellulae of 3 lengths; edge concolorous with the sides, smooth. Stipe 13–60 × 7–12 mm, equal, slightly attenuated towards apex, cylindric, slightly compressed at the apex, hollow, caespitose; surface uniformly orange-red (8B7), becoming deep orange (6A8) at maturity, often with light yellow (4A5) tints, smooth, dry. Context very thin, up to 2 mm thick at the disc, white.

Basidiospores dimorphous: macrospores ( $12.8 \pm 0.7 \times 7.8 \pm 0.7$ ) (11–) 11.5–13.5(–15) × 7–9(–10)  $\mu\text{m}$ ,  $Q=1.6$ , ellipsoid to elongate ellipsoid, hyaline, thin-walled with few refractive guttules; microspores ( $5.5 \pm 0.4 \times 3.4 \pm 0.2$ ) (4.5–) 5–6(–6.5) × 2.9–4  $\mu\text{m}$ ,  $Q=1.6$ , ellipsoid to elongate ellipsoid, similar to macrospores. Basidia dimorphous: macrobasidia 42.5–57 × 10–13  $\mu\text{m}$ , cylindric-clavate, bearing four thick, large sterigmata, 5.5–9.5 × 1.5–2.5  $\mu\text{m}$ , thin-walled, with numerous guttules; microbasidia 29–39 × 5.5–6.5  $\mu\text{m}$ , cylindric-clavate, bearing four sterigmata, up to 5.5  $\mu\text{m}$  long, similar to macrobasidia. Lamella-edge fertile. Cystidia absent. Hymenophoral trama regular, hyaline, of thin-walled hyphae, 1.5–7.5  $\mu\text{m}$  diam., inflated to 17.5  $\mu\text{m}$  diam. Subhymenial layer little developed, up to 8  $\mu\text{m}$  wide, loosely interwoven. Pileal context consisting of closely interwoven, thin-walled, hyaline hyphae, 1.5–5  $\mu\text{m}$  diam., inflated to 13  $\mu\text{m}$  diam.; oleiferous hyphae scattered, thick-walled, 2–7  $\mu\text{m}$  diam. Pileal surface a repent cutis of radially arranged parallel hyphae, 1.5–7.5  $\mu\text{m}$  diam., inflated to 22.5  $\mu\text{m}$  diam. Clamp-connections abundant.

HABITAT - On ground, caespitose, in wet evergreen tropical forest.

DISCUSSION: The characteristic features of *Hygrocybe manadukaensis* are the presence of deep yellow to deep orange or orange-red, smooth, convex pileus, light yellow to butter-yellow, subdecurrent lamellae, orange-red to deep orange, long and thick stipe, caespitose growth, and strongly dimorphic spores and basidia.

Among the varieties of *Hygrophorus firmus* described by Corner (1936), *Hygrocybe manadukaensis* closely resembles var. *militaris* and var. *puniceoides* in its similar sized and shaped macrospores. However, var. *militaris* clearly differs in having scarlet pileus and white stipe and var. *puniceoides* has a much larger (70–80 mm) pileus and longer (60–75 mm) stipe.

*Hygrocybe manadukaensis* more closely resembles *H. trinitensis* (Dennis) Pegler (Pegler 1983) in possession of a convex, shallowly depressed pileus and dimorphous basidiospores and basidia. However, *H. trinitensis* is clearly distinguished macroscopically by its small, scurfy, umbilicate pileus, coral-red lamellae, and thin, scarlet stipe and microscopically by its smaller (10–13 ×

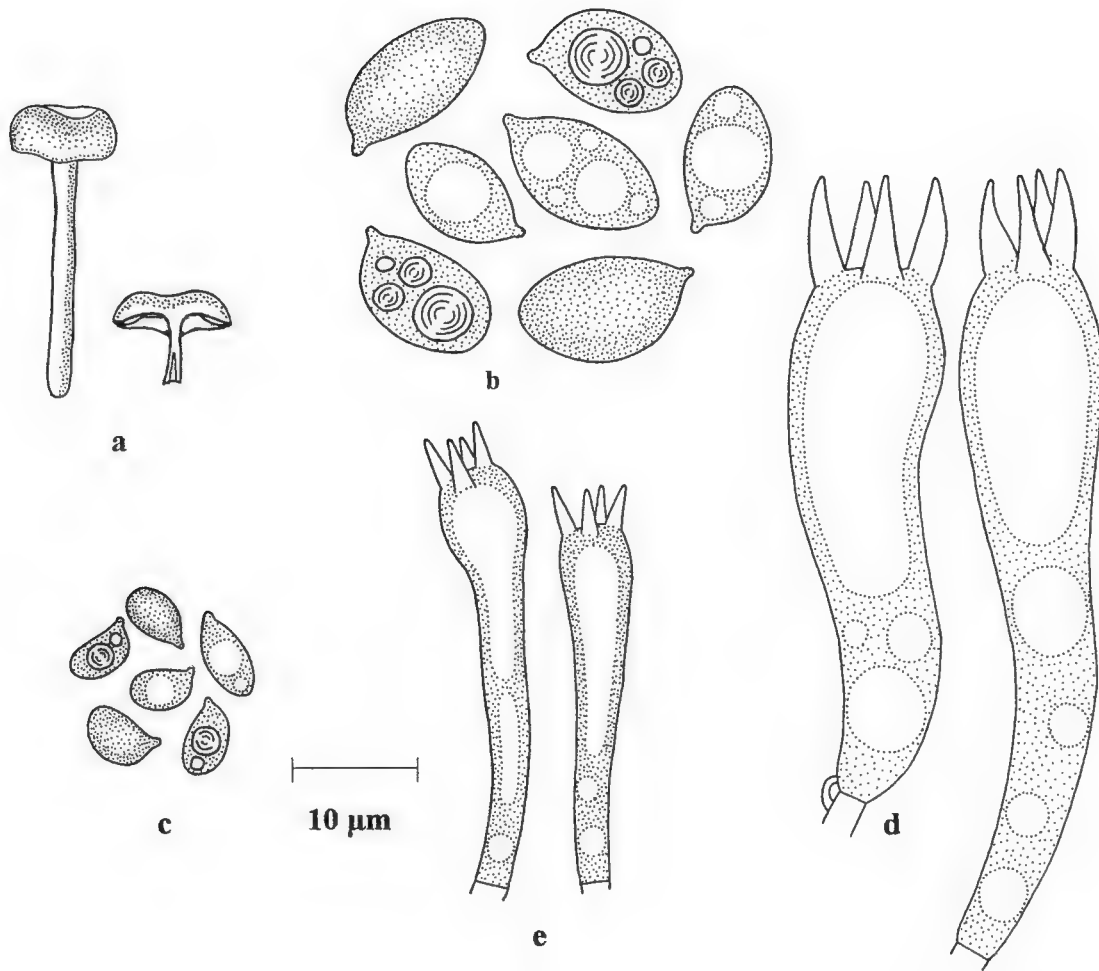


FIG 2. *Hygrocybe manadukaensis*: a. Habit  $\times 1$ . b. Macrobasidiospores, c. Microbasidiospores. d. Macrobasidia. e. Microbasidia. Scale bar = 10  $\mu\text{m}$ .

6–7.5  $\mu\text{m}$ ) macrospores, larger (7–9  $\times$  4.5–5.5  $\mu\text{m}$ ) microspores, and smaller macrobasidia (35–45  $\times$  8–9  $\mu\text{m}$ ) and microbasidia (20–28  $\mu\text{m}$ ).

*Hygrocybe occidentalis* (Dennis) Pegler var. *occidentalis* (Pegler 1983; Lodge & Pegler 1990) exhibits a similar range of yellow to orange colour variation and produces similarly sized macrospores and microbasidia. However, *H. occidentalis* var. *occidentalis* clearly differs macroscopically from *H. manadukaensis* in its larger (10–70 mm), convex to applanate, perforated pileus and larger (35–100  $\times$  4–20 mm) stipe; the latter species, which possesses a convex, depressed but never perforated pileus, is differentiated microscopically by its smaller microspores (5–8  $\times$  3.3–5  $\mu\text{m}$  in *H. occidentalis* var. *occidentalis*). In addition, both species differ in their growth habit, where *H. manadukaensis* produces caespitose basidiomes in contrast to the solitary to scattered habit of *H. occidentalis* var. *occidentalis*.

*Hygrocybe anisa* (Berk. & Broome) Pegler (Pegler 1986) produces similarly coloured and sized, caespitose basidiomes, macrospores, and microbasidia. However, *H. anisa* differs macroscopically from *H. manadukaensis* in its



straw yellow, slightly floccose/squamose pileus that lacks the orange tints that characterize *H. manadukaensis* and slender (2–5 mm) stipe. In addition, *H. anisa* is distinguished microscopically by larger microspores ( $6.5\text{--}8 \times 4.5\text{--}5.3 \mu\text{m}$ ) and macrobasidia (60–70  $\mu\text{m}$ ).

While the dimensions of the macro- and microspores of *H. natarajanii* are similar to those of *H. manadukaensis*, *H. natarajanii* has a yellow pileus covered with ruby red, tomentose squamules and a light yellow, longer, slender (50–140  $\times$  2–5 mm) stipe. In addition, *H. natarajanii* has larger macro- (55 – 68.5  $\mu\text{m}$ ) and micro- (37–44.5  $\mu\text{m}$ ) basidia (Senthilarasu et al. 2010).

*Hygrocybe manadukaensis* somewhat resembles *H. firma* (Berk. & Broome) Singer (Pegler 1986) in the orange to pale yellow convex pileus, subdecurrent pale yellow lamellae, long, thick, orange to pale yellow stipe, and similarly sized macrospores and microbasidia. However, *H. firma* clearly differs macroscopically in its tomentose to scurfy squamulose/fibrillose, perforated pileus, contrasting with the non-perforated smooth pileus of *H. manadukaensis*. In addition, *H. firma* microscopically differs in its larger microspores ( $6\text{--}8 \times 4.5\text{--}6 \mu\text{m}$ ) and macrobasidia (50–75  $\times$  12–16  $\mu\text{m}$ ).

The morphological variation observed in the specimen from Manaduka differentiates it from the above taxa and supports it as a new species in section *Firmae*.

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## MYCOTAXON

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***Coprinellus mitrinodulisporus*,  
a new species from chamois dung**FRANCESCO DOVERI\*, SABRINA SARROCCO, SUSANNA PECCHIA,  
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**Abstract** — The genus *Coprinellus* is re-examined from its establishment, through demotion as a synonym of *Coprinus*, and up through its current reinstatement. An agaric with a setulose pileus, sphaerocystic veil, and mitriform, nodulose spores has been isolated from chamois dung and, based on morphological data, is regarded as a new species in *Coprinellus*. The new taxon is compared with morphologically similar *Coprinellus* species, particularly with those having mitriform spores. Other taxa recently described in *Coprinus* are transferred to *Coprinellus*.

**Key words** — *Agaricales*, *Setulosi*, 28S rDNA, ITS,  $\beta$ -tubulin

**Introduction**

Persoon (1797) erected the genus *Coprinus* to accommodate agaric species with an ephemeral, membranous cap and blackening, deliquescent gills. Karsten (1879) later established the genus *Coprinellus* for species differing from *Coprinus* in having “caps covered by a cuticle or veil, finally lacerate and turned upwards” rather than “scaly from remnants of the universal veil, and covered by a veil”. Ricken (1915), who accepted *Coprinellus* as a subgenus of *Coprinus* limited to non-deliquescent species, restricted subgen. *Coprinus* to species with deliquescent gills. Lange (1938) reinstated *Coprinellus* at the genus level to include some non-deliquescent species, which Singer (1986) later placed in *Coprinus* subsect. *Setulosi* J.E. Lange. Singer (1986), whose conceptions were basic to the modern taxonomy of *Agaricales* Underw., regarded *Coprinellus* as a later synonym of *Coprinus*.

M. Lange (1952), who studied pileocystidiate species from different geographical origins morphologically and with interfertility tests, showed that

some species consisted of more than one cryptic, intersterile entity. Uljé & Bas (1991) monographed the *setulosi* at subsection level of section *Pseudocoprinus* (Kühner) P.D. Orton & Watling and included species with a hymenidermal cuticle and setulae on cap and stem, sometimes in association with veil remnants.

Molecular phylogenetic studies (Hopple & Vilgalys 1994, 1999; Johnson & Vilgalys 1998; Johnson 1999; Moncalvo et al. 2000, 2002) show *Coprinus comatus* (O.F. Müll.) Pers. (the type species of *Coprinus*) and its allies as distantly related to the other *Coprinus* species, and reveal *Coprinus* sensu lato to be a heterogeneous, polyphyletic assemblage. Based on these results, Redhead et al. (2001) split *Coprinus* s.l. into four genera — *Coprinus* s. str. in *Agaricaceae* Chevall. and *Coprinellus*, *Coprinopsis* P. Karst., and *Parasola* Redhead et al. in *Psathyrellaceae* Vilgalys et al. Their concept of *Coprinellus* includes species that in traditional systematics belong to subsect. *Setulosi* and subsects. *Domestici* Singer and *Micacei* (Fr.) Uljé & Noordel. of sect. *Veliformes* (Fr.) Penn. and are characterised by a hymenidermal or cystodermal pileipellis with a globular veil and/or pileocystidia (setulae).

This new taxonomy based on phylogenetic relationships in association with morphological features is now accepted by many authors, including Keirle et al. (2004) in their research on Hawaiian *Agaricales*, Nagy et al. (2009), who applied it to a complex study on *Parasola*, and Schafer (2010), who combined earlier subsections as sections of *Parasola*, *Coprinellus*, and *Coprinopsis*.

Since Uljé & Bas (1991) monographed *Setulosi*, additional new *Coprinus* s.l. species belonging to this section have been published (Uljé & Verbeken 2002, Uljé & Keizer 2003, Uljé & Noordeloos 2003, Nagy 2006), a few of which have been recombined in *Coprinellus* (Nagy et al., in press).

Our systematic study of coprophilous ascomycetes and basidiomycetes from Italy has recently allowed us to observe the growth on dung, in a damp chamber culture, of a *Coprinus* s.l., whose morphological features match those of *Setulosi*, but whose combination of characters does not correspond to any species in this section. We describe it here as a new species of *Coprinellus*.

## Materials and methods

### Isolation of the fungus — Morphological studies

Samples of chamois (*Rupicapra rupicapra*) dung were dried and cultured, after nineteen months, in a non-sterilised damp chamber according to Richardson & Watling (1997) and Richardson (2001), slightly modified by Doveri (2004). The cultures, placed under natural light at room temperature (18–25°C), were observed daily for five weeks with the unaided eye and a  $\times 7$ –45 magnification stereomicroscope. The macroscopic features were immediately described, and fresh material was mounted in water and Congo red and microscopically examined under a binocular light microscope. Spore



size was measured in water and calculated on 80 mature spores from 3 basidiomata, excluding the apiculum from the measurements (Q means the quotient of length divided by the breadth in face view). Small fruitbodies were dried in a few minutes with an artificial light. The collection has been preserved as dried material and slides (PI). Herbarium abbreviation follows Holmgren & Holmgren (1998).

### Molecular studies

DNA extraction was performed on a dried fruitbody using the DNeasy Plant MiniKit (Qiagen), according to the manufacturer's protocol. Polymerase Chain Reaction (PCR) was used to amplify the LSU and the ITS regions of the nuclear ribosomal DNA, employing the following primers: LR7, LR5, LR3R and LROR for the first 1.5 kb of the LSU gene and ITS1 and ITS4 for the ITS region (Gardes & Bruns 1993). Amplification reaction mixtures contained 25–50 ng of template DNA, GoTaq®Green Master Mix (Promega) 1X and 0.5 mM of each primer in a volume of 50 µL.

Amplification was performed in a GeneAmp® PCR System 2400 (Perkin Elmer) using the following parameters: for LSU initial denaturation step at 94°C for 5 min, 35 cycles consisting of denaturation at 94°C for 1 min, annealing at 50°C (for LROR/LR7) or 52°C (for LROR/LR5 and LR3R/LR7) for 1 min and extension at 72°C for 2 min, final extension of 72°C for 7 min; for ITS initial denaturation step at 94°C for 1 min, 30 cycles consisting of denaturation at 94°C for 30 s, annealing at 54°C for 1 min and extension at 72°C for 1 min, final extension of 72°C for 4 min. After the final extension of 72°C reactions were held at 4°C.

In addition, a fragment of the  $\beta$ -tubulin gene was amplified by primers B36f\_psa/B12r\_psa according to Nagy et al. (2010).

PCR products were purified by the QIAquick PCR Purification Kit (Qiagen) according to the manufacturer's protocol and submitted to sequencing. Samples to be sequenced were processed by the DNA Sequence Facility at the Bio Molecular Research (BMR), Servizio di Sequenziamento – CRIBI, University of Padova (Italy). For sequencing the same primers as described above for the ITS fragments and LR16 or LR22 as additional primers for the LSU fragment (Gardes & Bruns 1993) were used. The LSU and ITS sequences derived from these studies have been deposited in GenBank and compared with other sequences in GenBank.

### Taxonomy

#### *Coprinellus mitrinodulisporus* Doveri & Sarrocco sp. nov.

MYCOBANK 518735; GENBANK HQ180170

PLATE 1-2

*Pileus primo subglobosus vel elliptico-paraboliformis, usque ad 2 mm altus, deinde convexo-conicus vel campanulato-convexus, ultimo convexo-applanatus vel etiam revolutus, 3–10 mm latus, dense pruinoso-pubescent, radialiter striatus, deinde fissuratus. Cuticula primo ochracea, luteo vel purpureo, raro olivaceo, soffusa, deinde floris lactis colorem accipiens, plerumque ad marginem pallidior, ad postremum cinerascens. Lamellae ascendentes, stipitem non attinentes, ventricosae, infrequentes, ex albo nigricantes, ad marginem pallidiores. Stipes usque ad 45 × 0.5–0.8 mm, albidus vel modice quam pileus pallidior, flexuosus, cylindratus, aliquanto ad basim dilatatus at non bulbosus, omnino pruinoso-pubescent, saepe basali radiato mycelio praeditus. Inodorus. Velum granulatum, et ad*

*pileum et ad stipitem conspersum, ex crasse crustatis atque crassitunicatis sphaerocytibus, 12–20 µm diam., compositum. Sporae (9–) 9.5–11 (–11.5) × 6–7 × 5–6 µm, in adverso visu mitriformes, a latere subamygdaliformes, plerumque rotunde quadrinodosae, fuscobadiae, valde excentrico, 1.5–2 µm lato, poro germinativo praeditae. Basidia 16–27 × 6–9 µm, tetraspora, claviformia vel subcylindrata. Pleurocystidia absentia. Cheilocystidia copiosa, globosa vel late ellipsoidea, pedicularia, 25–39 × 21–32 µm. Pileipellis ex globosis, claviformibus vel late ellipsoideis, interdum crustatis cellulis, 20–48 × 16–34 µm, composita. Pileocystidia copiosa, lageniformia, et tenuitunicata, 62–78 (–90) × 12–15 µm, ad acutum apicem contracta, aliquando crustata leptocystidia, et crassitunicata, 35–45 × 11–16 µm, plerumque crustata sclerocystidia. Caulocystidia copiosa, pileocystidiis similia, 40–75 × 10–17 µm. Fibulae absentes. Holotypus hic designatus N.A. 1 in Pisani Horti Botanici viridario conservatur, ex fimo Rupicaprae rupicaprae, in Augustana Italica terra (saltus Salati) invento atque culto, ad viginti solitaria specimina remota, 28 Augustus 2008.*

TYPE: Salati pass (45°52'34"N 7°52'05"E), Aosta, Italy, on chamois dung, 28.8.2008, leg.: L. Levorato (Holotype N.A. 1, Pisa Botanical Garden)

ETYMOLOGY: *mitri-noduli-sporus* from the Latin (in turn from the Greek) “*mitra*” = “mitre”; “*nodulus*” = “small knob”; “*spora*” = “spore”, referred to the nodulose, mitriform spores

**MACROCHARACTERS**—PILEUS subglobose or ellipsoid-paraboloid when still closed, up to 2 mm high, convex-conic to conic-campanulate later, expanding to convex-plane or even revolute with an even margin, not umbonate, 3–10 mm diam., wholly and densely pruinose-pubescent, pruina thinning away with age, radially striate, becoming slightly grooved. Cuticle ochreous at first, with orange to purplish, rarely olive, shades, becoming cream coloured with a darker disc, finally greyish; LAMELLAE ascendant, free, ventricose, thin, distant, black at maturity with a paler edge. LAMELLULAE present; STIPE up to 45 × 0.5–0.8 mm, whitish or slightly paler than cap, wavy, cylindric, somewhat enlarged but not bulbous at the base, hollow, entirely pruinose-pubescent, often with a radial, white mycelial felt; VEIL granulose, present both on the cap and stem; CONTEXT imperceptible. No smell.

**MICROCHARACTERS**—BASIDIOSPORES (9–)9.5–11(–11.5) × 6–7 × 5–6 µm, mitriform in frontal view ( $Q = 1.38–1.69$ ;  $Q$  average = 1.52), subamygdaliform in side view, with a conical base and conical or convex apex, nodulose usually having two knobs on each side in face view, dark reddish brown at maturity, with a well developed, prominent apiculus, and an eccentric germ pore, 1.5–2 µm diam.; BASIDIA 4-spored, 16–27 × 6–9 µm, bimorphic, claviform or subcylindric, the latter with a slight median constriction, each surrounded by 4–5 globose to claviform brachybasidia, 17–33 × 17–30 µm.; PLEUROCYSTIDIA absent; CHEILOCYSTIDIA abundant, globose or broadly ellipsoidal, with a pedicel, 25–39 × 21–32 µm.; PILEIPELLIS a hymeniderm of globose, claviform, or broadly ellipsoidal, sometimes encrusted cells, 20–48 × 16–34 µm.; PILEOCYSTIDIA numerous, of two kinds, both lageniform: 1) thin-walled (leptocystidia), 62–78(–90) × 12–15 µm, bulbous at the base, with a neck tapering upwards,

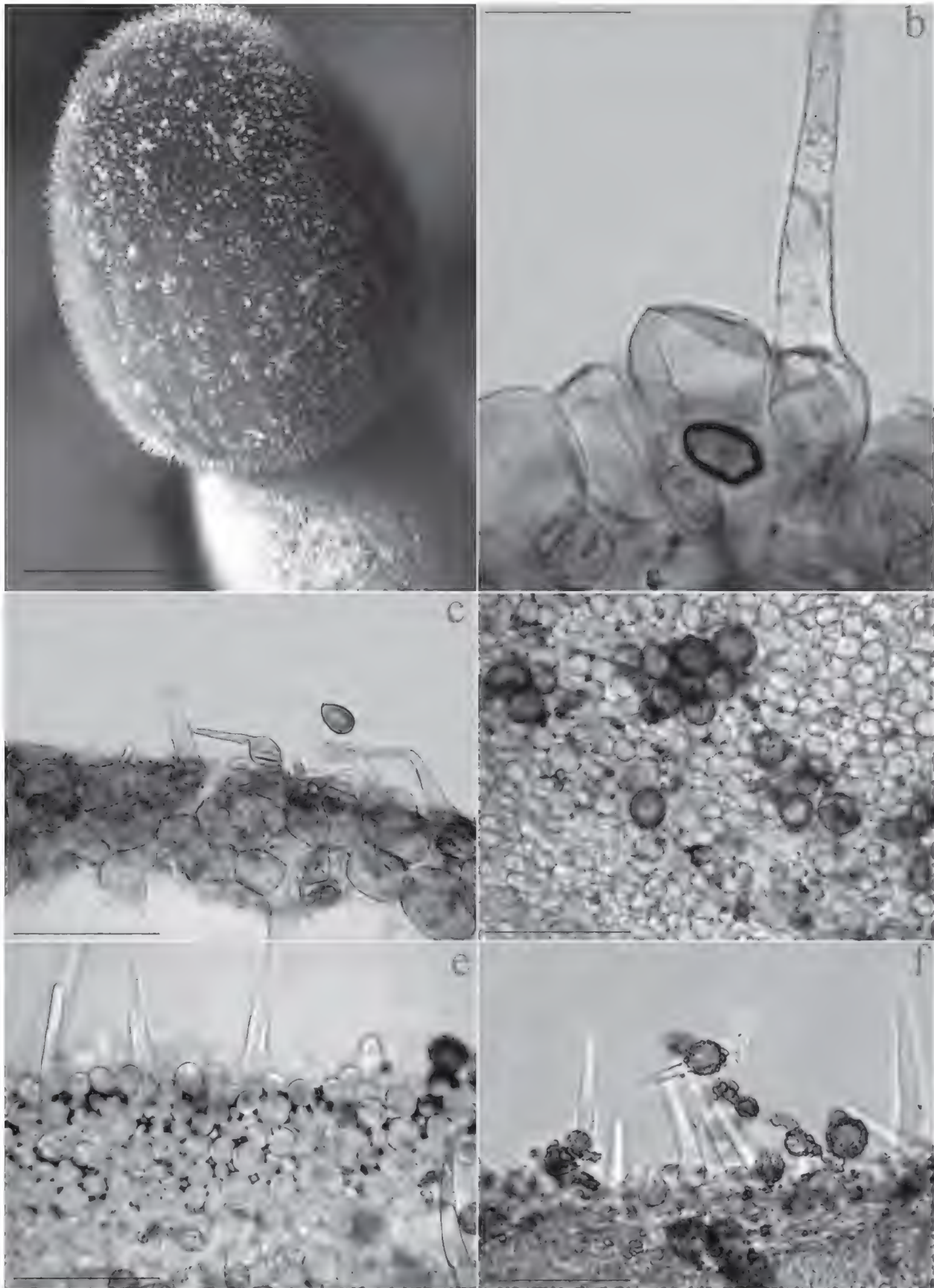


FIG. 1 *Coprinellus mitrinodulisporus* (holotype): a = basidioma in an early stage; b = hymenidermal cells interspaced with a leptopileocystidium; c, e–f = details of pileipellis with leptocystidia, sclerocystidia, and dark pigmented veil cells; d = dark pigmented veil cells above the hymenidermal cells. Scale bars: a = 500  $\mu$ m; b = 20  $\mu$ m; c–f = 50  $\mu$ m.



7–10 µm diam. at their base, sometimes sparsely encrusted at the neck and densely and coarsely at the base; 2) thick-walled (sclerocystidia), 35–45 × 11–16 µm (3–4 µm diam. at the neck base), darker than leptocystidia, usually coarsely encrusted at their bulbous base; CAULOCUTIS with the outermost hyphae 1–3 µm diam., sometimes encrusted, supporting many cystidia similar to pileocystidia, 40–75 × 10–17 µm.; VEIL formed of coarsely encrusted, thick-walled sphaerocysts, 12–20 µm diam., globose or even in transitional forms, with hints of neck, from sphaerocysts to sclerocystidia; CLAMP-CONNECTIONS absent.

ECOLOGY, RANGE, DISTRIBUTION—About twenty scattered specimens on chamois (*Rupicapra rupicapra*) dung in a damp chamber culture. August. To date only known from the type locality.

MOLECULAR ATTRIBUTES—Amplification of the LSU and ITS regions resulted in about 1.4 kb and 600 bp long sequences, respectively. Comparison of our LSU sequence (accession number HQ180170) with those deposited in GenBank resulted in high similarity percentages (96%) with other strains of *Coprinellus* spp., and comparison of the ITS sequence (accession number HQ180171) within the same database confirmed this result. A β-tubulin sequence has been deposited (HQ180172) to support further phylogenetic studies on *C. mitrinodulisporus*.

## Discussion

The main features of *Coprinellus mitrinodulisporus* are growth on dung, pileus with setulae, and a granulose, sphaerocystic veil, the latter particularly evident in the early stages, mitriiform and nodulose basidiospores, and absence of clamp-connections. The presence of a hymenidermal pileipellis and setuliform pileo- and caulocystidia places the species in subs. *Setulosi* of Uljé & Bas (1991) and now in *Coprinellus*, as revised and reinstated by phylogenetic studies (Redhead et al. 2001) as section *Setulosi* (J.E. Lange) D.J. Schaf. (Schafer 2010).

*Coprinellus mitrinodulisporus* is very close to *Coprinus doverii* L. Nagy, a typical representative of *Setulosi* not yet recombined in *Coprinellus* (Nagy, in litt.). The two species share habitat and many macro- and microscopic features, including encrusted lageniform pileocystidia and mitriiform nodulose spores, but *C. mitrinodulisporus* differs in having larger spores (6.2–8.3 × 4.5–5.8 × 3.8–4.1 µm in *C. doverii*), abundant and larger cheilocystidia (gill edge almost sterile), longer pileocystidia, abundant sclerocystidia and veil (the latter easily observable with a ×10 magnification), and in lacking clamp connections, which are absent also in the mycelial felt. In addition, *C. mitrinodulisporus* has pileocystidia with constantly tapering necks rather than with both tapering and cylindrical necks. Given the limited number of collections of both species

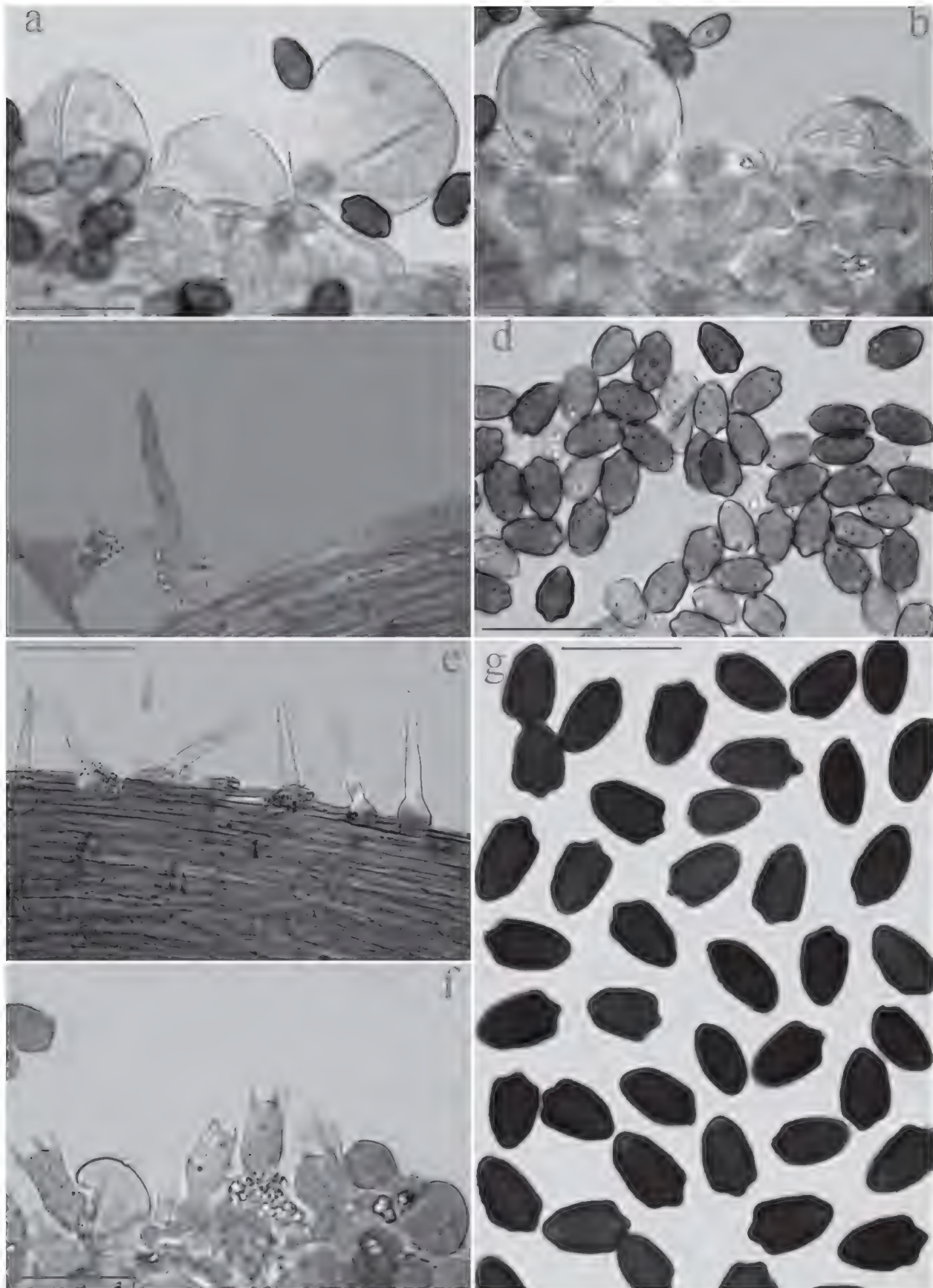


FIG. 2 *Coprinellus mitrinodulisporus* (holotype): a = brachybasidia; b = cheilocystidia; c, e = details of caulocutis with lageniform cystidia; d = immature and maturing spores; f = detail of hymenium with basidia; g = mature spores. Scale bars: a–d, f = 20  $\mu$ m; e = 50  $\mu$ m; g = 15  $\mu$ m.



studied and the unique combination of characters they share, they might conceivably represent one, variable taxon. Further studies are clearly desirable but considering the observed differences and the possible rarity of the taxa, we prefer to treat these as separate species. As the two species occupy an isolated morphological position in *Setulosi*, a molecular phylogeny might not clearly differentiate them from each other. It would be interesting to explore their intersterility in mating studies.

Lageniform leptopileo- and scleropileocystidia tapering upwards and similarly sized ( $8.5\text{--}11 \times 6.5\text{--}8.5 \times 5\text{--}6.5 \mu\text{m}$  in Orton & Watling 1979), mitriform basidiospores are also found in *Coprinellus angulatus* (Peck) Redhead et al., which is, however, a carbonicolous species with much larger, rust-brown fruitbodies, non-nodulose, more squat basidiospores (average length/breadth =  $1.25\text{--}1.35$ , Uljé 2005) with a central, very wide and truncate germ pore, pleurocystidia, and clamp-connections.

*Coprinellus marculentus* (Britzelm.) Redhead et al., a coprophilous pileocystidiate species with a granular veil and similarly sized basidiospores also shares purplish pileus shades and globose or broadly ellipsoidal cheilocystidia (Uljé & Bas 1991), but *C. marculentus* differs in its smooth, usually hexagonal, sometimes mitriform basidiospores, and pileocystidia with a cylindrical neck, equal or enlarged at its apex. It also differs from *C. mitrinodulisporus* in lacking sclerocystidia and having pleurocystidia and clamp-connections.

Although it does not have mitriform spores, *Coprinellus heptemerus* (M. Lange & A.H. Smith) Vilgalys et al. has other characters in common with *C. mitrinodulisporus*, including an encrusted veil with cells transitional between sphaerocysts and pileocystidia, a lack of clamp connections and pleurocystidia, small fruitbodies, and a habit on dung. However, the combination of characters and distinctly shaped spores distinguish *C. mitrinodulisporus* clearly from *C. heptemerus* and other previously published *Setulosi*, except *C. doverii*.

Apart from *C. doverii*, no other *Setulosi* species published after Uljé & Bas (1991) and Uljé & Noordeloos (2003) has coarsely encrusted veil sphaerocysts, sclerocystidia and mitriform basidiospores, easily distinguishing them from *C. mitrinodulisporus*. We take the opportunity to recombine some of them in *Coprinellus*:

***Coprinellus allovelus* (Uljé) Doveri & Sarrocco, *comb.nov.***

MYCOBANK 518736

= *Coprinus allovelus* Uljé, in Uljé & Noordeloos, Persoonia 18: 261, 2003

***Coprinellus limicola* (Uljé) Doveri & Sarrocco, *stat. nov., comb.nov.***

MYCOBANK 518737

= *Coprinus callinus* var. *limicola* Uljé, in Uljé & Noordeloos, Persoonia 18: 259, 2003 as "*limicolus*"

NOTE: Nagy (pers. comm.) reports that, based on molecular results, this is a separate species. Morphologically it has a number of differences from *C. callinus* that support its rank as a distinct species. M. Lange (1952), who reported that collections identified morphologically as *C. callinus* consisted of two intersterile taxa, was not able to distinguish these morphologically.

*Coprinellus canistri* (Uljé & Verbeken) Doveri & Sarrocco, **comb.nov.**

MYCOBANK 518738

≡ *Coprinus canistri* Uljé & Verbeken, Persoonia 18: 143, 2002

*Coprinellus minutisporus* (Uljé) Doveri & Sarrocco, **comb.nov.**

MYCOBANK 518739

≡ *Coprinus minutisporus* Uljé in Uljé & Noordeloos, Persoonia 18: 260, 2003

*Coprinellus pseudoamphithallus* (Uljé) Doveri & Sarrocco, **comb.nov.**

MYCOBANK 518741

≡ *Coprinus pseudoamphithallus* Uljé in Uljé & Noordeloos, Persoonia 18: 263, 2003

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## MYCOTAXON

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**A new species of *Phlyctis* (*Phlyctidaceae*) from China**RUI MA<sup>1</sup>, HONG-MEI LI<sup>2</sup>, HAI-YING WANG<sup>1\*</sup> & ZUN-TIAN ZHAO<sup>1\*</sup>*lichenmr@yahoo.com* \* *lichenwhy@yahoo.com.cn* \* *ztzhao@sohu.com*<sup>1</sup>*College of Life Sciences, Shandong Normal University  
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**Abstract** — A new *Phlyctis* species, *P. subargena*, characterized by a sorediate thallus, clustered apothecia and 2-spored asci, is described from north-central China.

**Key words** — lichen, ascomycetes, Asia, taxonomy

**Introduction**

After Flotow (1850) established the lichen genus *Phlyctis* (Wallr.) Flot., the genus was expanded to include taxa formerly placed in *Phlyctomia*, *Phlyctella*, and *Phlyctidia* (Galloway & Guzmán 1988). Following phylogenetic analyses of molecular data, *Phlyctis* was moved from the *Lecanorales* to the *Ostropales* (Wedin et al. 2005, Miadlikowska et al. 2006). *Phlyctis* species are morphologically characterized by crustose thalli; small innate or subimmersed apothecia; large, colourless, and septate or muriform ascospores, 1–2 or 8 per ascus; and globose green algae as photobionts (Purvis et al. 1992, Brodo et al. 2001, Tønsberg 2004, Galloway 2007). *Phlyctis* species contain one or several of the following depsidone acids: stictic, constictic, norstictic, connorstictic, hypostictic, salazinic, psoromic, neopsoromic, and protocetraric (Galloway & Guzmán 1988).

*Phlyctis* contains approximately 12 species worldwide (Kirk et al. 2008), but only *Phlyctis schizospora* Zahlbr., from Hubei Province, has been reported from China (Chen et al. 1989, Wei 1991). During our study of *Phlyctis* collected from Gansu Province, an interesting *Phlyctis* species new to science was found.

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## Materials and methods

The specimens studied were collected from Gansu Province, China, and are preserved in SDNU (Lichen Section of Botanical Herbarium, Shandong Normal University). The morphology of the lichen specimens was examined using a stereo microscope (COIC XTL7045B2) and a compound microscope (JNOEC XS-213). Lichen substances in all specimens cited were identified using the standardized thin layer chromatography techniques (Culberson 1972). Photos of the thallus and ascospores were taken under OLYMPUS SZX12 with DP70.

## Taxonomy

*Phlyctis subargena* R. Ma & H.Y. Wang, sp. nov.

FIG. 1

MYCOBANK 518778

*Species acido norstictico, sporis 2nae et sorediis copiosis a congeneribus diversa.*

TYPE COLLECTION: CHINA. Gansu province, Longnan, Wenxian Co. Qiujiaba, alt. 2450m, on bark, F. Yang, 20070050, 2 August 2007. (Holotype in SDNU).

EXPANDED DESCRIPTION —Thallus crustose, 60–120 µm thick, distinctly sorediate; surface arachnoid-bysoid, forming patches, roughened-uneven to irregularly areolate; areolate 0.1–0.2 mm, greenish white; prothallus white at margins and breaks in thallus; soralia usually paler than thallus, powdery to granular, coalescing to form diffuse, irregular patches. Apothecia frequent, 0.1–0.3 mm in diam, 3–8(–10) clustered, immersing in thalline sorediate patches; disc reddish-brown, rounded to irregularly, plane, usually with white pruina; exciple poorly developed. Epihymenium yellow-brown, up to 30 µm thick; hymenium colourless, up to 130 µm thick, hypothecium pale to light brown, up to 30 µm thick; paraphyses slender, simple; asci broadly clavate, 110–150 × 32–40 µm, 2-spored; ascospores hyaline, muriform, 42–78 × 30–42 µm; I–. Photobiont green, globose, 12–18 µm in diam.

CHEMISTRY — Cortex K+ yellow, C–; medulla K+ yellow-orange-red, C–, PD+ yellow. Constituent in 6 specimens tested: norstictic acid.

SUBSTRATE AND DISTRIBUTION —*Phlyctis subargena* is a corticolous species, found only in the type locality at present.

ADDITIONAL SPECIMENS EXAMINED —CHINA. Gansu: Longnan, Wenxian Co., Qiujiaba, alt. 2450m, on bark, 2/VIII/2007, F. Yang 20070024, 20070043, 20070045; alt. 2350m, on bark, 3/VIII/2007, F. Yang 20070080; alt. 2350m, on bark, 5/VIII/2007, F. Yang 20070381, 20070383-1(SDNU).

COMMENTS —The presence of norstictic acid, abundant soredia, and two spores per ascus distinguishes *Phlyctis subargena* from all other *Phlyctis* species. *Phlyctis agelaea* (Ach.) Flot., *P. chilensis* D.J. Galloway & Guzmán, *P. oleosa* Stirt., *P. speirea* G. Merr., *P. uncinata* Stirt. and *P. argena* (Ach.) Flot.



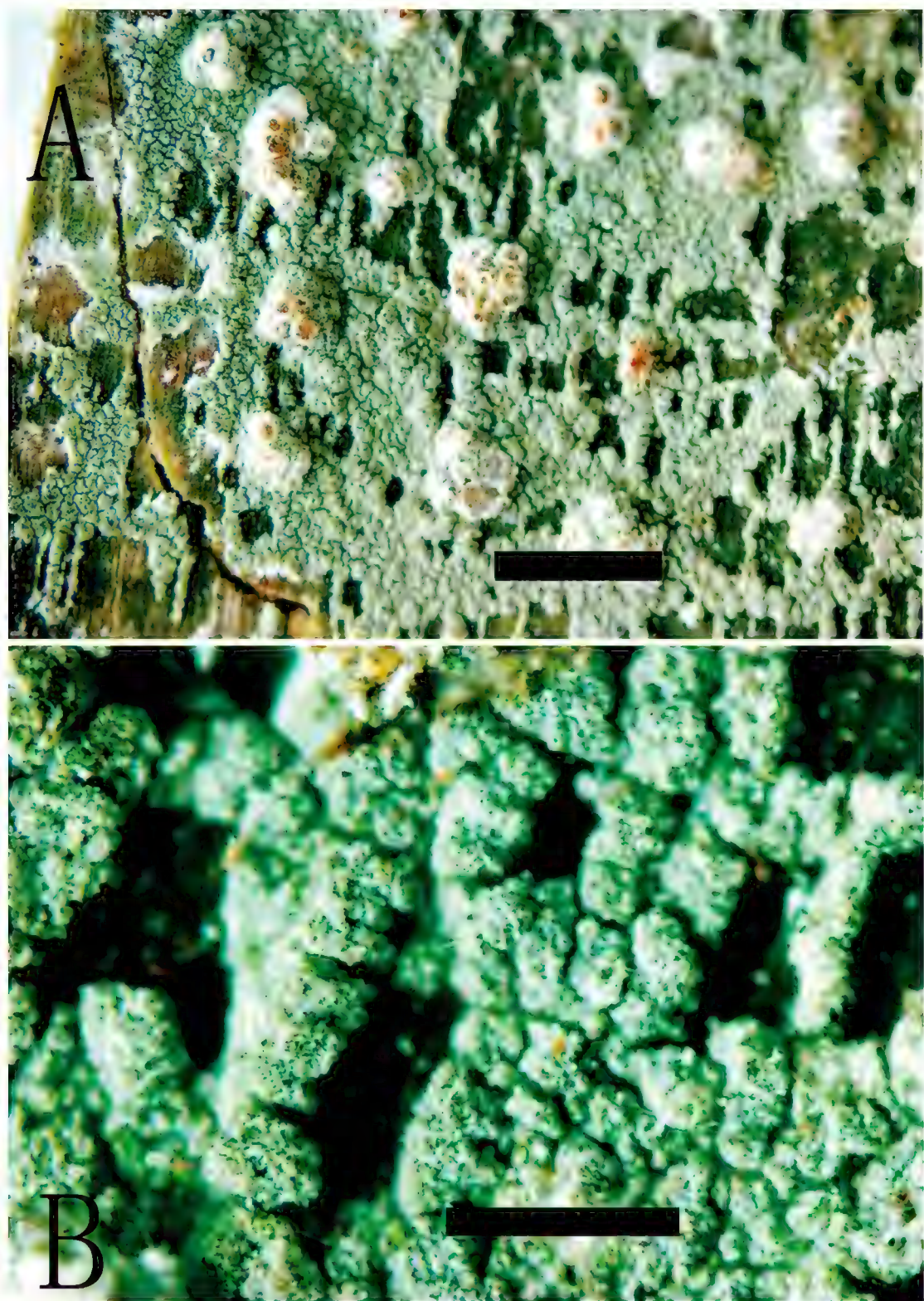


FIG. 1. *Phlyctis subargena* (holotype). A. Thallus (bar = 2 mm). B. Soralia (bar = 200  $\mu$ m).

all contain norstictic acid. However, the former five are esorediate. Although *P. argena* is distinctly soorediate, *P. subargena* can be clearly separated from the former, which produces rare and solitary apothecia, only one spore per ascus, and larger spores ( $100\text{--}150 \times 25\text{--}50 \mu\text{m}$ ). In addition, *P. argena* also contains a trace of connorstictic acid, which is absent in *P. subargena*.





FIG. 1. *Phlyctis subargena* (holotype). A. Apothecium (bar = 50  $\mu$ m).  
B. Ascospores, showing 2-spored ascus and muriform shape (bar = 20 $\mu$ m).

*Phlyctis subuncinata* Stirt., which is also sorediate, differs from *P. subargena* in its fusiform spores and chemistry (stictic and cryptostictic acid vs. norstictic acid).

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## MYCOTAXON

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**Two new species of *Kylindria* from Fujian, China**

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**Abstract** — Two new species of *Kylindria* were found during a survey of anamorphic fungi in tropical areas of Fujian province, China. The new species, *K. millettiae* and *K. embeliae*, occurred on the hosts *Millettia championii* and *Embelia rudis*, respectively. They are described, illustrated, and compared with closely related taxa. The type specimens are deposited in HSAUP (Herbarium of the Department of Plant Pathology, Shandong Agricultural University) and HMAS (Mycological Herbarium, Institute of Microbiology, Chinese Academy of Sciences).

**Key words** — hyphomycetes, taxonomy

**Introduction**

The genus *Kylindria* was erected by DiCosmo et al. (1983) based on *Cylindrotrichum triseptatum* Matsush. (Matsushima 1975). In a revision of the species of *Cylindrotrichum* Bonord. and *Chaetopsis* Grev., five species were assigned to the new genus *Kylindria*. The distinguishing characters of *Kylindria* were considered to be the macronematous, mononematous, dark, conidiophores, the monophialidic, narrow conidiogenous cells, and aseptate or one to several septate, smooth, hyaline conidia usually with an eccentric protruding basal hilum (DiCosmo et al. 1983, Castañeda 1988). These characters separate the genus from similar genera such as *Cylindrotrichum*, *Xenokylindria* DiCosmo et al., and *Chaetopsis* (DiCosmo et al. 1983).

Up to now, the genus *Kylindria* contains 13 species, and no species have been reported from China. In our studies on hyphomycetes from deciduous stems and rotten wood in south of China, two previously undescribed species of *Kylindria* were found. They are proposed herein as new.

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Taxonomy

*Kylindria millettiae* Y.D. Zhang & X.G. Zhang, sp. nov.

FIG 1

MYCOBANK MB 518820

*Coloniae effusae, brunneae, pilosae. Mycelium partim superficiale et partim immersum, ex hyphis ramosis, septatis, laevibus, pallide brunneis, 2.5–3 µm crassis compositum. Conidiophora macronematosa, mononematosa, nonramosa, erecta, recta vel flexuosa, laevia, atro-brunnea, apice versus pallidiora, 7–10-septata, 220–265 µm longa, 5.5–7.5 µm*



FIG. 1. *Kylindria millettiae* A. Conidiophores with conidia. B. Conidia.

*crassa. Cellulae conidiogenae monophialidica, cylindrica vel leviter subulata, integratae, terminales, dilute brunnea, 10.5–17 µm longa, 4.5–5.5 µm crassa, apicem versus deminutae. Conidia solitaria, cylindrica, hyalina, laevia, 3-septata, in massis mucosis translucentibus formata, apicem obtusa, 19.5–24 µm longa, 6.5–9 µm crassa.*

HOLOTYPE: on dead branches of *Millettia championii* Benth. (Leguminosae), forest park of Wuyishan, Fujian Province, China, 16 Aug. 2009, Y.D. Zhang, HSAUPH3023 (isotype HMAS 146114).

ETYMOLOGY: in reference to the host genus, *Millettia*.

Colonies effuse, brown, hairy. Mycelium partly superficial, partly immersed, composed of branched, septate, smooth-walled, pale brown hyphae, 2.5–3 µm thick. Conidiophores macronematous mononematous, unbranched, erect, straight or flexuous, smooth, dark brown, paler towards the apex, 7–10 septate, 220–265 µm long, 5.5–7.5 µm wide. Conidiogenous cells monophialidic, cylindrical or tapered, integrated, terminal, pale brown, 10.5–17 µm long, 4.5–5.5 µm wide, narrower at the apex. Conidia solitary, cylindrical, hyaline, smooth, 3-septate, accumulating in translucent slimy masses at the apices of conidiogenous cells, 19.5–24 µm long, 6.5–9 µm wide, obtuse at the apex, with an excentric, lateral, flat scar on the second cells from base.

The conidia of *K. millettiae* are morphologically similar to those of *K. excentrica* Bhat & B. Sutton (Bhat & Sutton 1985) in conidium morphology. However, the conidia of *K. millettiae* are smaller than those of *K. excentrica* (19.5–24 × 6.5–9 µm vs. 27.5–35 × 7.5–8.5 µm). In addition, most conidia of *K. millettiae* have an excentric lateral flat scar arising from the second cells close to base, whereas *K. excentrica* produces a lateral flat scar on the basal cells of the conidia.

***Kylindria embeliae* Y.D. Zhang & X.G. Zhang, sp. nov.**

FIG 2

MYCOBANK MB 518821

*Coloniae effusae in substrato naturali, olivaceo-brunneae vel fuscae, pilosae. Mycelium partim superficiale et partim immersum, ex hyphis ramosis, septatis, pallide brunneis vel brunneis, laevibus, 1.5–2.5 µm crassis compositum. Conidiophora macronematosa, mononematosa, nonramosa, erecta, recta vel flexuosa, laevia, atro-brunnea, apice versus pallidiora, 5–7-septata, 130–150 µm longa, 5.5–6.5 µm crassa. Cellulae conidiogenae monophialidica, cylindrica, integratae, ad subapicem inflatae, 15–19.5 µm longa, 6.5–7.5 µm crassa, cum collaretto cupulato. Conidia solitaria, ellipsoidea vel cylindrica, hyalina, laevia, aseptata, 17.5–23 µm longa, 6–7.5 µm crassa, apice rotundata, ad basim truncata.*

HOLOTYPE: on dead branches of *Embelia rudis* Hand.-Mazz. (Myrsinaceae), forest park of Wuyishan, Fujian Province, China, 15 Aug. 2009, Y.D. Zhang, HSAUPH3007 (isotype HMAS 146115).

ETYMOLOGY: in reference to the host genus, *Embelia*.

Colonies effuse on the natural substratum, olivaceous brown to blackish brown, hairy. Mycelium partly superficial and partly immersed composed of branched, septate, pale brown to brown, smooth-walled hyphae, 1.5–2.5 µm thick.



FIG. 2. *Kylindria embeliae* A. Conidiophores with conidia. B. Conidia.

Conidiophores macronematous, mononematous, unbranched, erect, straight or flexuous, smooth, dark brown, paler towards the apex, 5–7-septate, 130–150 µm long, 5.5–6.5 µm wide. Conidiogenous cells monophialidic, cylindrical, integrated, swollen at the subapical region, 15–19.5 µm long, 6.5–7.5 µm wide, occasionally with a collarette at the apex. Conidia solitary, ellipsoidal or cylindrical, hyaline, smooth, aseptate, 17.5–23 µm long, 6–7.5 µm wide, apex rounded, base truncate.

Four other described species of *Kylindria* have aseptate conidia — *K. conglutinata* Matsush. (Matsushima 1993), *K. obesispora* R.F. Castañeda

(Castañeda 1988), *K. keitae* Rambelli & Onofri (Rambelli & Onofri 1987), *K. peruamazonensis* Matsush. (Matsushima 1993), and *K. zignoellae* (Höhn.) DiCosmo et al. (DiCosmo et al. 1983). The conidia of *K. embeliae* are larger than those of *K. keitae* ( $17.5\text{--}23 \times 6\text{--}7.5 \mu\text{m}$  vs.  $12.5\text{--}16.5 \times 4.5\text{--}5.5 \mu\text{m}$ ). In addition, the conidiogenous cells of *K. embeliae* become swollen at the subapical region and occasionally possess a collarete.

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## MYCOTAXON

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**A new species of *Minimelanolocus* from Fujian, China**

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**Abstract** — *Minimelanolocus chimonanthi* sp. nov. is described and illustrated occurring on dead branches of *Chimonanthus nitens*. The specimen was collected from tropical forests in Fujian Province of China. The type specimen is deposited in HSAUP (Herbarium of the Department of Plant Pathology, Shandong Agricultural University) with an isotype in HMAS (Mycological Herbarium, Institute of Microbiology, Chinese Academy of Sciences).

**Key words** — anamorphic fungi, taxonomy

**Introduction**

Castañeda & Heredia established the genus *Minimelanolocus* based on 12 previously described species of *Pseudospiropes* M.B. Ellis, *Helminthosporium* Link, and *Belemnospora* P.M. Kirk with *M. navicularis* (R.F. Castañeda) R.F. Castañeda as the type species. The generic characteristics of *Minimelanolocus* include macronematous, mononematous, dark conidiophores, holoblastic, polyblastic, indeterminate, terminal becoming intercalary, integrated conidiogenous cells with holoblastic sympodial extensions and inconspicuous or slightly prominent, narrow, opaque, refractive to somewhat obscure dehiscence scars, and euseptate conidia; conidial secession is schizolytic (Castañeda et al. 2001).

To date, of the 18 taxa of *Minimelanolocus* accepted worldwide, most are saprobes on rotten leaves or dead twigs, dead wood, and bark. Five species (*M. endospermi*, *M. pterocarpi*, *M. magnoliae*, *M. machili*, *M. camelliae*) have been reported from China (Ma et al. 2008, Zhang et al. 2009). A survey of the saprobic fungi on dead wood from tropical forest in Fujian Province of China has revealed a previously undescribed species of *Minimelanolocus*. It is proposed herein as new.

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FIG. 1. *Minimelanolocus chimonanthi*  
A. Conidiophores conidiogenous cells with conidia. B. Conidia.

## Taxonomy

***Minimelanolocus chimonanthi*** Y.D. Zhang & X.G. Zhang, sp. nov.

FIG 1

MYCOBANK MB 518829

*Coloniae effusae in substrato naturali, brunneae, pilosae. Mycelium partim superficiale et partim immersum, ex hyphis ramosis, septatis, pallide brunneis, laevibus, 2–3 µm crassis compositum. Conidiophora macronematosa, mononematosa, solitaria, nonramosa, erecta, recta vel flexuosa, laevia, atro-brunnea, apice versus pallidiora, 5–10-septata, 160–250 µm longa, 6.5–10.5 µm crassa, circa apicem 5.5–6.5 µm crassa. Cellulae conidiogenae holoblasticae, polyblasticae, in conidiophoris incorporatae, indeterminatae, sympodialiter extendentes, terminales deinde intercalares, pallide brunneae. Loci conidiogeno inconspicuo vel leviter prominentibus, subobscuris. Conidia late fusiformia, breviter rostrata ad apicem, hyalina, solitaria, acropleurogena, simplicia, brunneae, laevia, 5–7-euseptata, 26–35 µm longa, 6.5–10 µm crassa. Conidiorum secessio schizolytica.*

HOLOTYPE: on dead branches of *Chimonanthus nitens* Oliv. (*Calycanthaceae*), forest park of Wuyishan, Fujian Province, China, 16 Aug. 2009, Y.D. Zhang, HSAUP H3002 (isotype HMAS 146111).

ETYMOLOGY: in reference to the host genus, *Chimonanthus*.

Colonies effuse on natural substratum, brown, hairy. Mycelium partly superficial, partly immersed, composed of branched, septate, pale brown, smooth-walled hyphae, 2–3 µm thick. Conidiophores macronematous, mononematous, unbranched, erect, straight or flexuous, smooth, dark brown, paler towards the apex, 5–10-septate, 160–250 µm long, 6.5–10.5 µm thick, near the apex 5.5–6.5 µm thick. Conidiogenous cells polyblastic, integrated, indeterminate, sympodial, terminal becoming intercalary, pale brown. Conidiogenous loci inconspicuous or slightly prominent. Conidia broadly fusiform, shortly rostrate at the apex, hyaline, solitary, acropleurogenous, simple, brown, smooth-walled, 5–7-euseptate, 26–35 µm long, 6.5–10 µm thick in the broadest part. Conidial secession schizolytic.

The conidia of *M. chimonanthi* are similar to those of *M. navicularis* (Castañeda et al. 2001). However, the conidia of *M. chimonanthi* are hyaline and larger than those of *M. navicularis* (26–35 × 6.5–10 µm vs. 20–25 × 6–8 µm). In addition, the conidia of *M. chimonanthi* are 5–7 septate while those of *M. navicularis* are only 3 septate.

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## **Austro-American lignocellulolytic basidiomycetes (*Agaricomycotina*): new records**

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**Abstract**— A survey of lignolytic basidiomycetes from Mondaí (27°06'16"S, 53°24'07"W) in the Brazilian state of Santa Catarina has revealed nine previously unrecorded species: *Dacryopinax elegans*, *Cotylidia aurantiaca*, *Hymenochaete rubiginosa*, *Inonotus rickii*, *Phellinus rhytiphloeus*, *Echinoporia aculeifera*, *Oxyporus obducens*, *Amauroderma sprucei*, and *Pseudofavolus miquelii*. Comments about the species and illustrations are provided.

**Key words** — mycodiversity, *Agaricomycetes*, *Dacrymycetes*

### **Introduction**

Among the estimated 1.5 million fungal species, only 74,000 to 120,000 species have been described. With limited human and financial resources, a total inventory is not possible within any reasonable time frame, which is estimated to be 1290 years at the current rate (Garibay-Orijel et al. 2009). Within the field of mycology, there are numerous studies about the diversity of macrofungi. However, Gilbert & Souza (2002) and Piepenbring (2007) point out that a significant portion of the fungal taxa from tropical forests has not yet been described.

In the southern region of coastal South America, the Atlantic Forest is broadly defined and includes not only coastal rain forests but also inland forests and coastal seasonal forests, which are mostly semi-deciduous and mixed *Araucaria* forests (Fernandes & Bezerra 1990).

Knowledge about the abundance of lignolytic basidiomycetes in all forest types, as well as the fact that they are the largely responsible for decaying wood in most ecosystems, is well established. However, fundamental questions, such as how many species are from a specific region or whether fungal diversity is greater in one forest type versus another, remain unanswered due to taxonomic



issues and the deficiency of long-term studies in many regions (Groposo et al. 2005). There is a common belief that some wood-decaying basidiomycetes generally have low host- and habitat-specificity, and this assumption somewhat complicates evaluation of the ecological specialization and species distribution based on past studies (Gilbert et al. 2008).

Regardless of its biological richness, the Atlantic Forest is probably one of the most highly threatened tropical forests in the world (Jarenkow & Budke 2009). In the past, commercial exploitation of this area has led to deforestation. Currently the Atlantic forest is extremely fragmented and many endemic species are endangered (Metzger 2009).

In the state of Santa Catarina, several studies have been published that include data about collections from the Atlantic Forest of Santa Catarina Island. However, in other areas of the state little is known about their mycodiversity. The work presented here — a result of the first extensive survey carried out in the deciduous seasonal forest of Santa Catarina — aims to expand the knowledge about the region's mycodiversity. It is also part of a current taxonomic and biogeographical survey of wood-inhabiting basidiomycetes in this state. Additional collections made during this survey from the municipality of Mondaí (from deciduous seasonal forest) resulted in several previously unrecorded species of *Agaricomycotina*, which are briefly discussed below.

## Material and methods

The municipality of Mondaí is located in the extreme western part of the state of Santa Catarina (27°06'16"S, 53°24'07"W), in Southern Brazil. Collections were made periodically between December 2005 and May 2007 at two locations (Linha Uruguai and Linha Sanga Forte) in Mondaí.

Macro- and microscopic data of the specimens were collected following traditional methodology (Singer 1975, Ryvarden 1991). Measurements were made from slide preparations stained with 1% phloxine solution + 1% or 5% KOH solution. Melzer's reagent was used to detect the presence of amyloid or dextrinoid reactions on the cell walls. Collections were identified by consulting literature and specimens in the following herbaria: BAFC, FLOR, ICN, NYBG, SP, URM (Holmgren & Holmgren 2009). Voucher specimens are stored at FLOR. Taxonomic arrangement follows Kirk et al. (2008).

## Taxonomy

*Dacryopinax elegans* (Berk. & M.A. Curtis) G.W. Martin, Lloydia 11(2): 116. 1948.

FIG. 1

≡ *Guepinia elegans* Berk. & M.A. Curtis, Hook. J. Bot. Kew Gdn Misc. 1: 239. 1849.

DESCRIPTION: McNabb (1965).

VOUCHER MATERIAL: BRAZIL. Santa Catarina: Mondaí, Linha Sanga Forte, Campos-Santana & Santana 302, 25/V/07 (FLOR 32214).

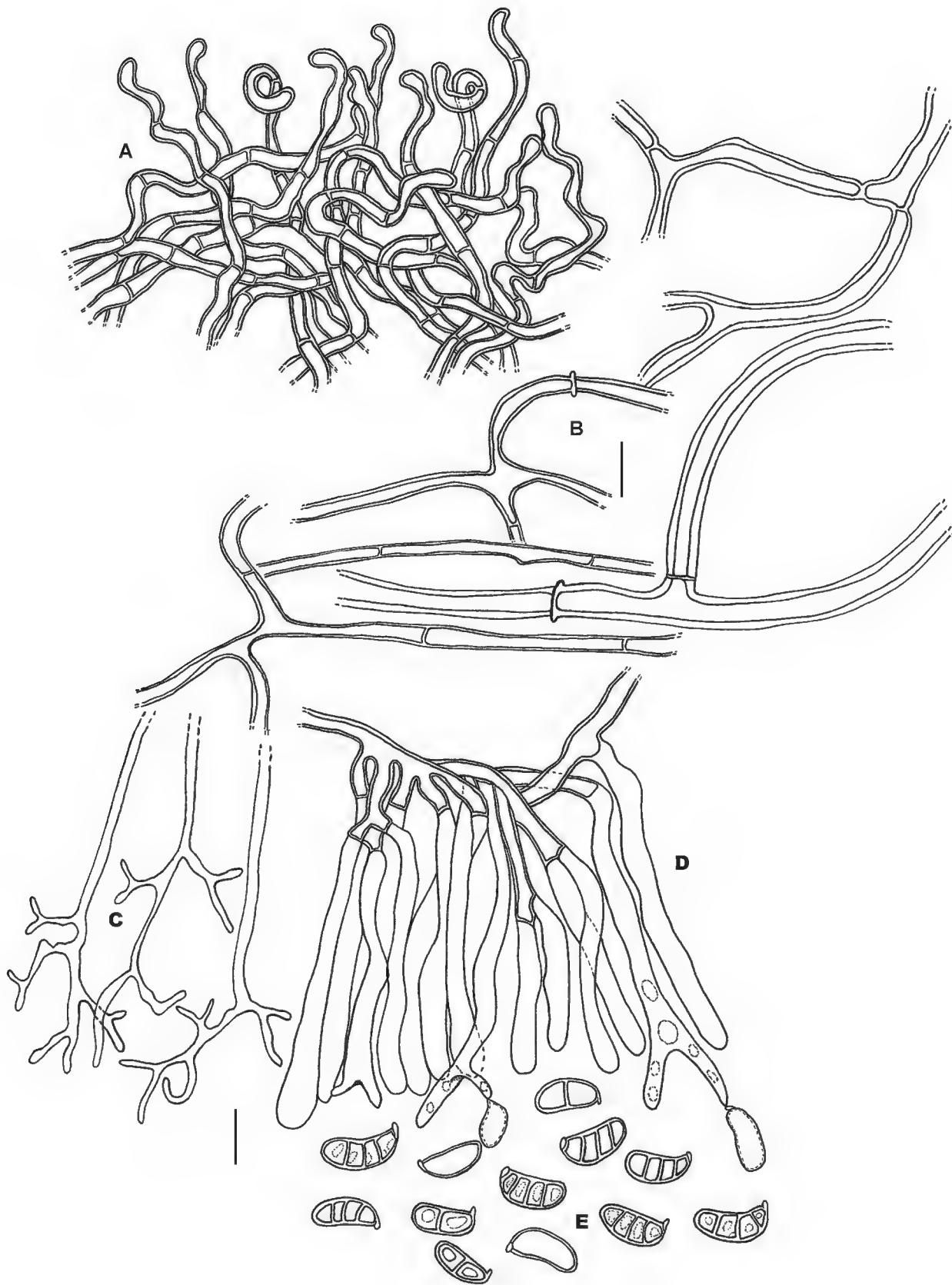


FIG. 1. *Dacryopinax elegans*. Scale = 10  $\mu$ m.

A. Septate hairs. B. Generative hyphae. C. Dendrohyphidium. D. Hymenium. E. Basidiospores.

COMMENTS: The species is recognized by its stipitate basidiomata, solitary or in groups; pileus spathulate, flabelliform, initially cupulate or obliquely cupulate;

consistency gelatinous or cartilaginous. Microscopically it is characterized by the presence of a cortex, medulla, and hymenium; cortex and stipe present cylindrical, tortuous, thin or thick walled, tinted brown septate hairs. The cylindrical-subclavate basidia with basal septa, become bifurcate and the basidiospores are cylindrical, thick-walled with thick septa, yellowish brown, apiculate, becoming 3-septate at maturity are characteristic. As noted by McNabb (1965), *D. elegans* is distinguished as the only *Dacryopinax* species with thick-walled hyphae and tri-septate basidiospores. In our collection, the basidiospores ( $12\text{--}14(-15) \times 5\text{--}6(-6.5) \mu\text{m}$ ) are similar to those observed by McNabb (1965;  $(12\text{--})14\text{--}15.5 \times 5\text{--}6.5 \mu\text{m}$ ) and Fonseca et al. (2002;  $13.6\text{--}15.6(-16) \times 5.6\text{--}6.4 \mu\text{m}$ ). However, López & Garcia (2001) cite slightly larger basidiospores ( $(13\text{--})14\text{--}16(-19) \times 5.6\text{--}6.04 \mu\text{m}$ ).

ADDITIONAL MATERIAL: ARGENTINA, Bs. As., Llava llo, Sta. Cat. Inst. Fitotéc., R.T.Guerrero, 18/IV/1963 (BAFC 23086); ibid, Sgo. del Estero, Depto Choya, el Salvador, R.E.dela Sota (Det. R.T. Guerrero), 20/V/1961 (BAFC 23097).

DISTRIBUTION: Brazil (Espírito Santo, Amazonas, Rio Grande do Sul, Rio de Janeiro, Roraima), Colombia, Costa Rica, Dominican Republic, Guiana, Jamaica, Mexico, Panama, Puerto Rico, Trinidad & Tobago, Venezuela (McNabb 1965, Fonseca et al. 2002, Roberts 1996, Sobestiansky 2005).

*Cotylidia aurantiaca* (Pers.) A.L. Welden, Lloydia 21: 40, 1958.

FIG. 2

= *Thelephora aurantiaca* Pers., Voy. Uranie, Bot. 5: 176, 1827.

DESCRIPTION: Reid (1965)

VOUCHER MATERIAL: BRAZIL, Santa Catarina: Mondaí, Linha Uruguai, Campos-Santana & Santana 205, 23/V/2007 (FLOR 32308); ibid, Linha Sanga Forte, Campos-Santana & Santana 262, 25/V/2007 (FLOR 32309).

COMMENTS: *Cotylidia aurantiaca*, which is one of the most common species collected in the tropical America (Reid 1965), exhibits a wide morphological variation, commonly spathulate, ligulate, flabellate or reniform, pseudo-infundibuliform or infundibuliform. This species is characterized by a bright yellow fresh hymenial surface that discolours to creamy-ochre when dry, basidiospores that are thin-walled, hyaline, elliptical, a monomitic hyphal system, and variably shaped cystidia, some of which develop 1–3 transverse septa and frequently constrict somewhat at these points. In our collection, the basidiospores ( $6\text{--}9 \times (2.5\text{--})4.5(-5) \mu\text{m}$ ) are similar to those observed by Reid (1965;  $(5.5\text{--})6\text{--}8.75(-9) \times 3\text{--}3.75(-4) \mu\text{m}$ ) and in one collection from Argentina (BAFC 24989:  $7\text{--}9 \times 2.5\text{--}4 \mu\text{m}$ ).

ADDITIONAL MATERIAL: ARGENTINA, Misiones, Colônia Belgrano, monte al SE próximo de la Estación Forestal, Wright, Deschamps & Del Busto, M-2455, 29/X/1973 (BAFC 24989).

DISTRIBUTION: Brazil (Rio de Janeiro, Amazonas, Rio Grande do Sul), Argentina, Costa Rica, Colombia, China, Ecuador, Paraguay, Santo Domingo, Trinidad (Dai et al. 2004, Reid 1965).

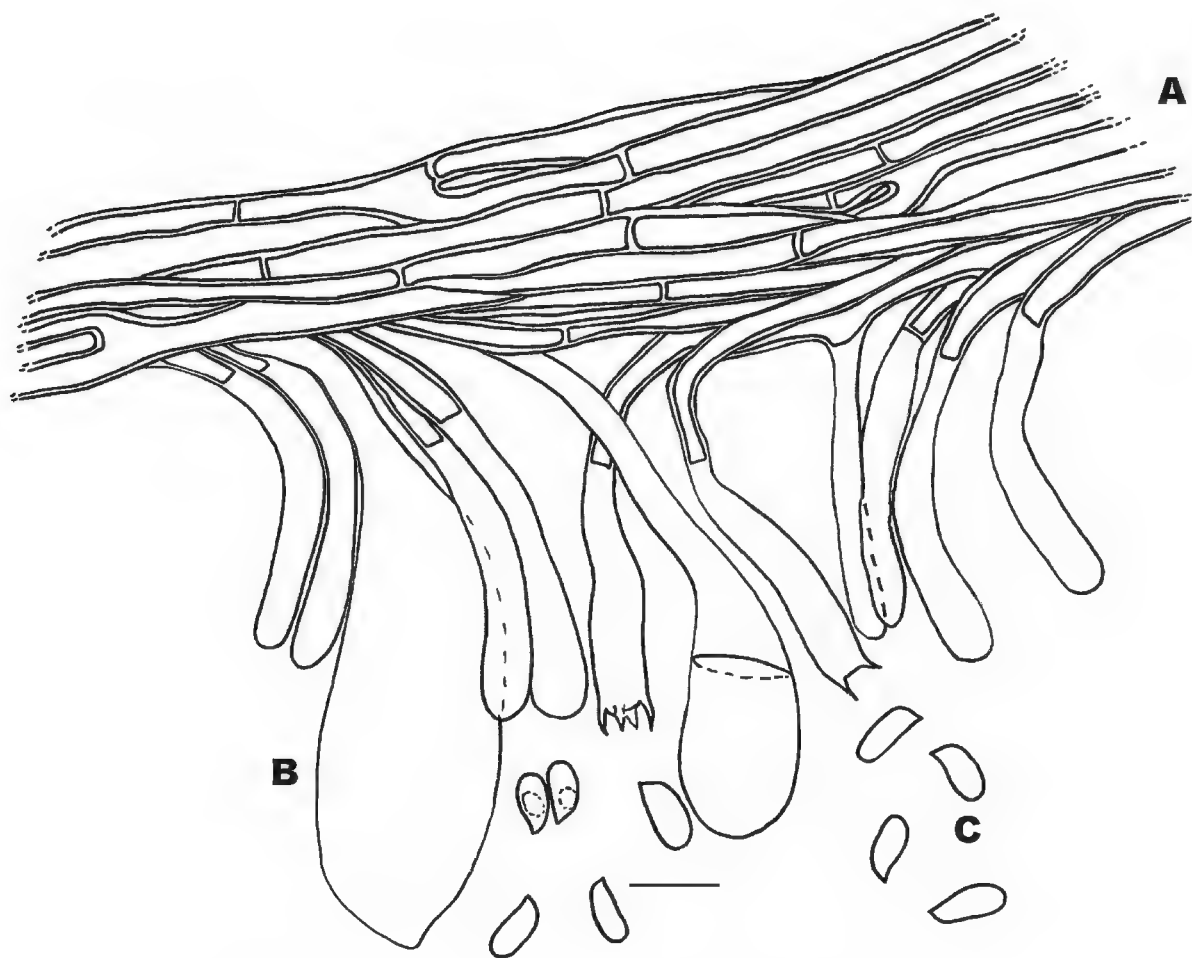


FIG. 2. *Cotylidia aurantiaca* hymenium. Scale = 10  $\mu\text{m}$ .  
A. Generative hyphae. B. Cystidia. C. Basidiospores.

*Hymenochaete rubiginosa* (Dicks.) Lév., Ann. Sci. Nat. Bot., 3e Sér., 5: 151, 1846.

FIG. 3

$\equiv$  *Helvella rubiginosa* Dicks. Fasc. Pl. Crypt. Brit. 1: 20, 1785.

DESCRIPTION: Job (1985)

VOUCHER MATERIAL: BRAZIL, Santa Catarina: Mondaí, Linha Sanga Forte, Campos-Santana, Santana & Rodrigues-Souza 10, 03/I/06 (FLOR 32215).

COMMENTS: The examined material is typical for this species. Basidiospore measurements ( $3\text{--}6 \times 2\text{--}2.5 \mu\text{m}$ ) were close to those recorded by Parmasto (2001;  $(3.5\text{--})3.8\text{--}5.5 \times (1.8\text{--})2\text{--}2.8\text{--}(3) \mu\text{m}$ ), and slightly smaller than those reported by Cunningham (1956;  $5.5\text{--}7 \times 3.5\text{--}4 \mu\text{m}$ ). This species is easily recognized in the field by its rigid reflexed margin, dark brown upper surface, and light yellowish brown to yellow hymenophore. Chamuris (1988) and Cunningham (1956) point out that these features distinguish *H. rubiginosa* from *H. tabacina* (Sowerby) Lév., which has a reflex flexible region, orange-brown upper surface, and pale hymenophore. Job (1985) observed that *H. rubiginosa* is one of the few species of the genus with a cosmopolitan distribution.



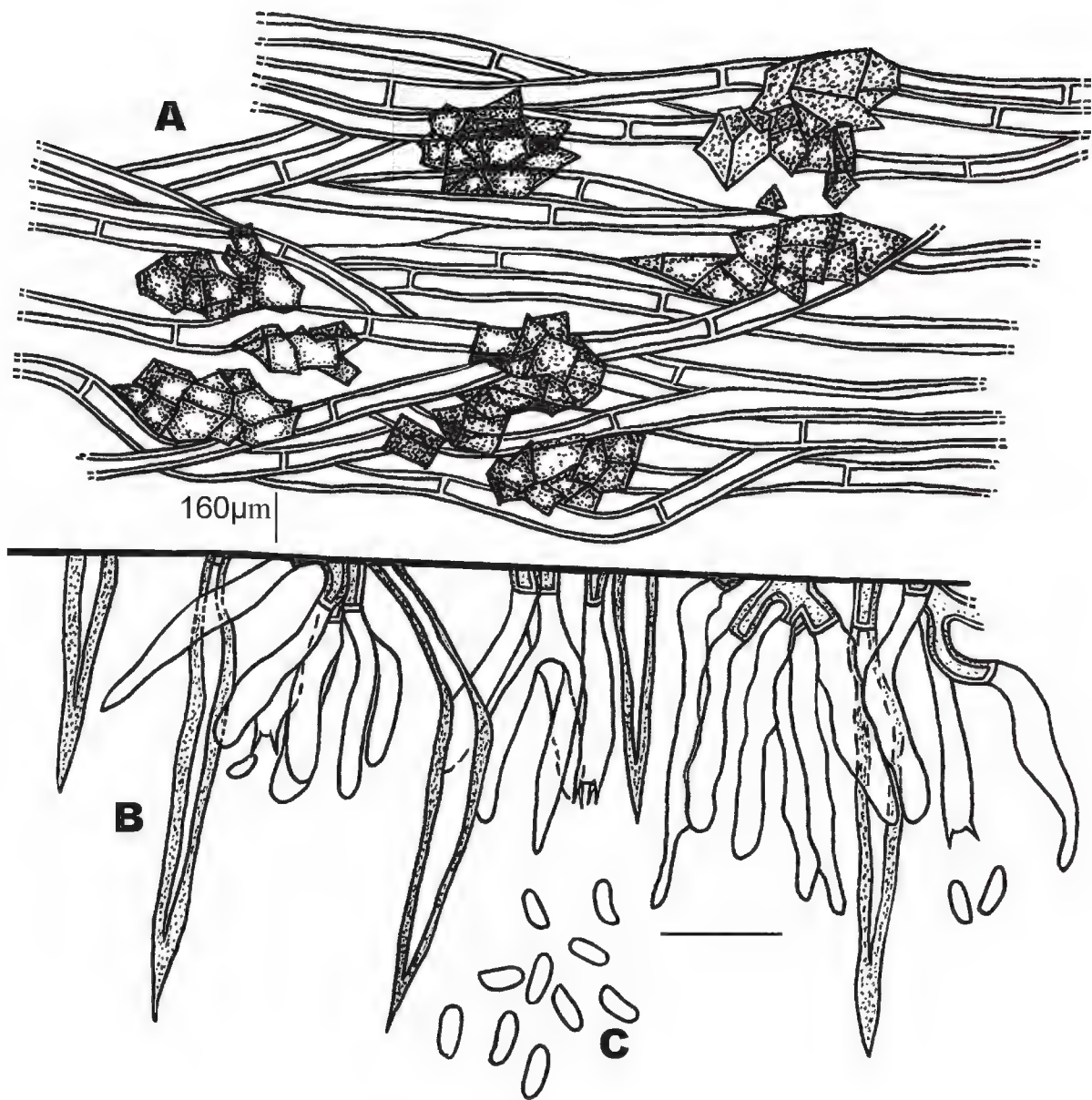


FIG. 3. *Hymenochaete rubiginosa*. Scale =10 µm.  
A. Context generative hyphae. B. Setae. C. Basidiospores.

ADDITIONAL MATERIAL: BRAZIL, São Paulo: Santo André, Reserva Biológica do Alto da Serra de Paranapiacaba, Trufem SB & Grandi RAP, 09/VIII/88 (SP 307428).

DISTRIBUTION: Cosmopolitan; Brazil (Rio Grande do Sul and São Paulo), Europe, North America, New Zealand, Norway, Central America and Argentina (Cunningham 1963, Fonsêca 1999, Job 1985, Reeves & Welden 1967, Ryvarden 1971).

*Inonotus rickii* (Pat.) D.A. Reid, Kew Bull. 12: 141, 1957.  
= *Xanthochrous rickii* Pat., Bull. Soc. Mycol. France 24(1): 6, 1908.

DESCRIPTION: Ryvarden (2005).

VOUCHER MATERIAL: BRAZIL, Santa Catarina: Mondaí, Linha Sanga Forte, Campos-Santana & Santana 288, 25/V/07 (FLOR 32216).

FIG. 4



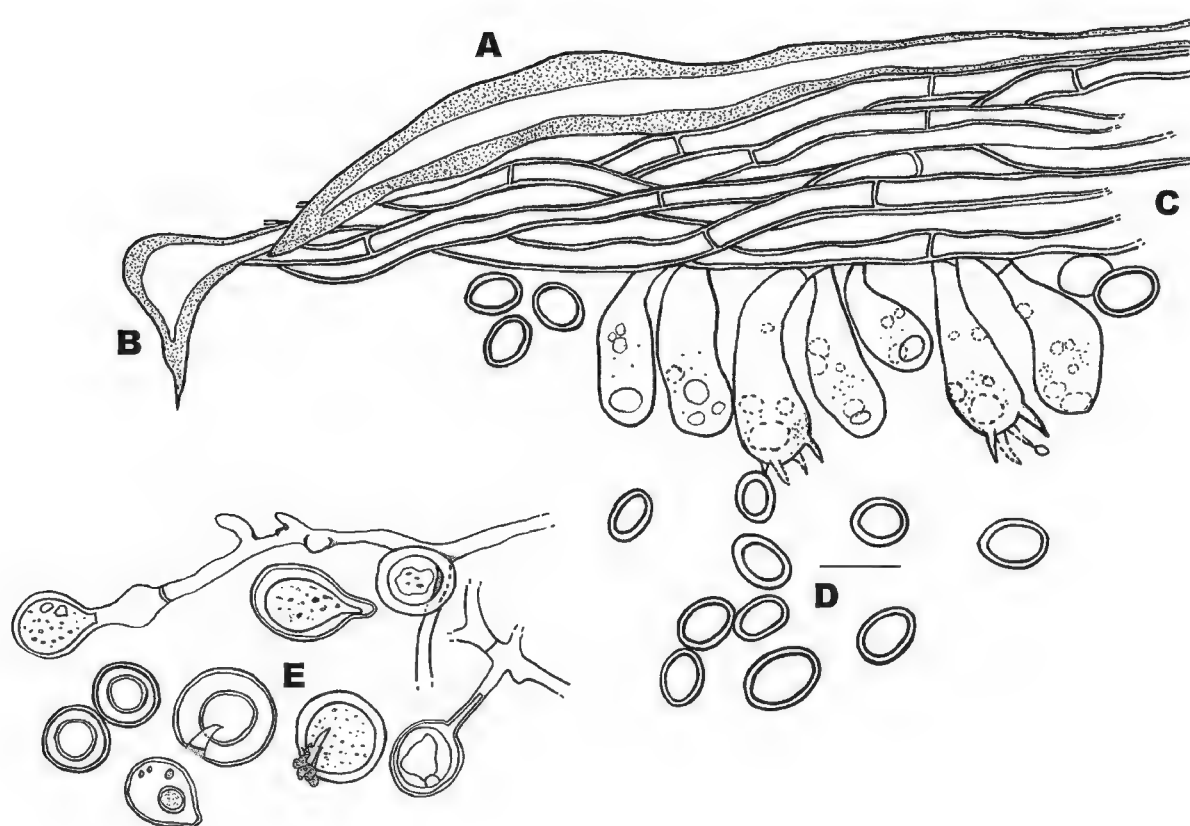


FIG. 4. *Inonotus rickii*. Scale = 10  $\mu\text{m}$ .

A. Setal hyphae. B. Hymenial setae. C. Generative hyphae.  
D. Basidiospores. E. Chlamydospores.

COMMENTS: Some authors, such as Coelho (1994), Melo et al. (2002), and Ryvarden (2005) described setal hyphae that ranged from  $250 \times 17.94 \mu\text{m}$ . Although these measurements are similar to those in the Mondaí specimens, the longer hyphal setae found in the context —  $400(-500) \times 9-20(-22) \mu\text{m}$  — agrees with the sizes cited reported by Intini & Tello (2003). Basidiospore size in our specimens ( $6-8 \times 4-7 \mu\text{m}$ ) is similar to that reported by Coelho (1994;  $6.55-8.95 \times 5.7-6.2 \mu\text{m}$ ) but larger than those reported by Melo et al. (2002) and Gilbertson & Ryvarden (1986;  $6-8.5(-9) \times 4.5-5.5 \mu\text{m}$ ). Abundant chlamydospores ( $8-18 \times 8-17 \mu\text{m}$ ) were found in the context, as observed by Melo et al. (2002).

ADDITIONAL MATERIAL: BRAZIL, Rio Grande do Sul: Porto Alegre, Ponta Grossa, Eny C. Vianna, IV/93 (ICN 97681); *ibid*, Parque da Redenção, R.T. Guerrero, I/90 (ICN 97594); *ibid*, Santa Maria, Itaara, Parque Pinhal, G. Coelho 24-13, 07/VI/1992 (ICN 97677); *ibid*, Caturrita, S. Aldorindo, G. Coelho 20-06, 1992 (ICN 97676).

DISTRIBUTION: Pantropical—North America, Central America, South America (Brazil in Rio Grande do Sul, Argentina), (Coelho 1994, Robledo & Rajchenberg 2007).

*Phellinus rhytiphloeus* (Mont.) Ryvar den, Prelim. Polyp. Fl. E. Africa: 206, 1980.

FIG. 5

≡ *Polyporus rhytiphloeus* Mont., Ann. Sci. Nat., Bot., 4e Sér., 5: 369, 1857.

DESCRIPTION: Ryvar den & Johansen (1980).

VOUCHER MATERIAL: BRAZIL, Santa Catarina: Mondaí, Linha Uruguai, Campos-Santana, Santana & Zanella 77, 15/VI/2006 (FLOR 32218); ibid, Campos-Santana & Santana 257, 290, 25/V/07 (FLOR 32219, FLOR 32220).

COMMENTS: Our specimens show 7–9 pores per mm and basidiospores measuring 4–5(–5.5)  $\mu\text{m}$  in diameter, as previously reported by Ryvar den & Johansen (1980). Globose, golden to rusty brown basidiospores and absence of setae are characteristic. As observed by Gibertoni (2004), basidiospore size and color and basidioma morphology distinguish *P. rhytiphloeus* from the other *Phellinus* species that lack setae. In their original description, Ryvar den & Johansen (1980) noted that the absence of setae differentiates *P. rhytiphloeus*

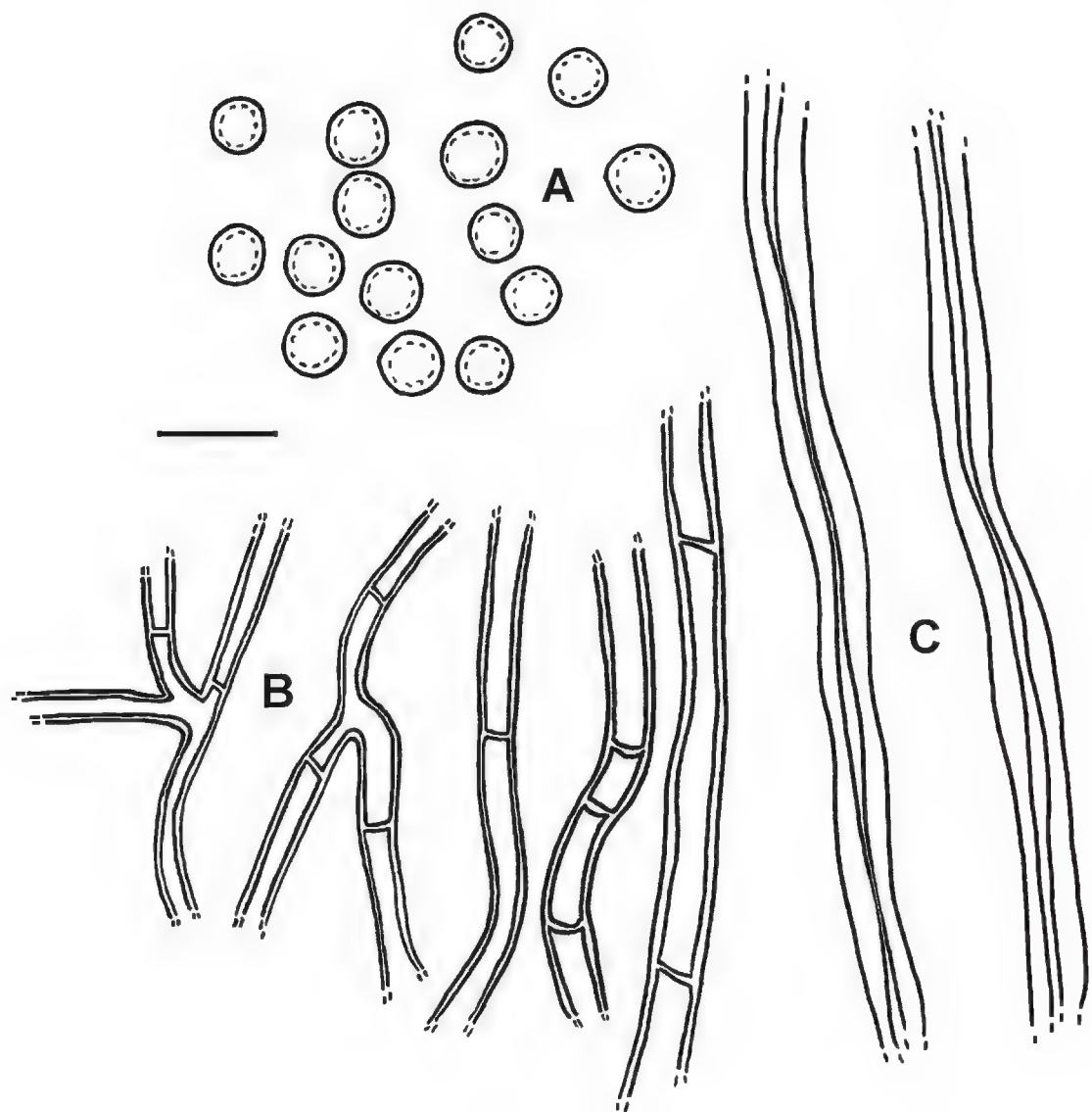


FIG. 5. *Phellinus rhytiphloeus*. Scale = 10  $\mu\text{m}$ .  
A. Basidiospores. B. Generative hyphae. C. Skeletal hyphae.

from *Phellinus rhabarbarinus* (Berk.) G. Cunn. (Gerber & Loguercio-Leite 1997). Our examinations of *P. rhabarbarinus* specimens (FLOR 10.922; FLOR 10.929) confirm this and also show that the size ( $3\text{--}4 \times 2\text{--}2.5\ \mu\text{m}$ ) of the hyaline ellipsoid basidiospores is another character that differentiates these species.

ADDITIONAL MATERIAL: BRAZIL, Rio Grande do Norte: Baía Formosa, RPPN Senador Antônio Faria-Mata Estrela, Gibertoni, V/20002 (URM 77794); *ibid*, Santa Catarina: Florianópolis, Morro da Lagoa da Conceição, Furlani & Loguercio-Leite, 186, 26/XII/1988 (FLOR 10929); *ibid*, Gerber & Cabral, 318, 12/XI/1993 (FLOR 10922); *ibid*, Willerding, A. & Santos, B., 420, 02/IV/94 (FLOR 10920); *ibid*, Santo Amaro da Imperatriz, Atanazio, J. & Willerding, A., 450, 20/V/1994 (FLOR 10928); *ibid*, Palhoça, Parque Estadual Serra do Tabuleiro-Cambirela, Groposo & Andrade, 176, 18/VII/2001 (FLOR 11957).

DISTRIBUTION: Neotropical; Brazil (Rio Grande do Norte), Jamaica, Surinam, Mexico and Venezuela (Gibertoni & Cavalcanti 2003, Ryvarden & Guzmán 1993, Ryvarden & Iturriaga 2001).

*Echinoporia aculeifera* (Berk. & M.A. Curtis) Ryvarden, Mycotaxon 20(2): 330, 1984.

FIG. 6

= *Trametes aculeifera* Berk. & M.A. Curtis, J. Linn. Soc., Bot. 10: 319, 1868.

DESCRIPTION: Silveira & Guerrero (1991).

VOUCHER MATERIAL: BRAZIL, Santa Catarina: Mondaí, Linha Uruguai, Campos-Santana & Santana 244, 23/V/07 (FLOR 32222).

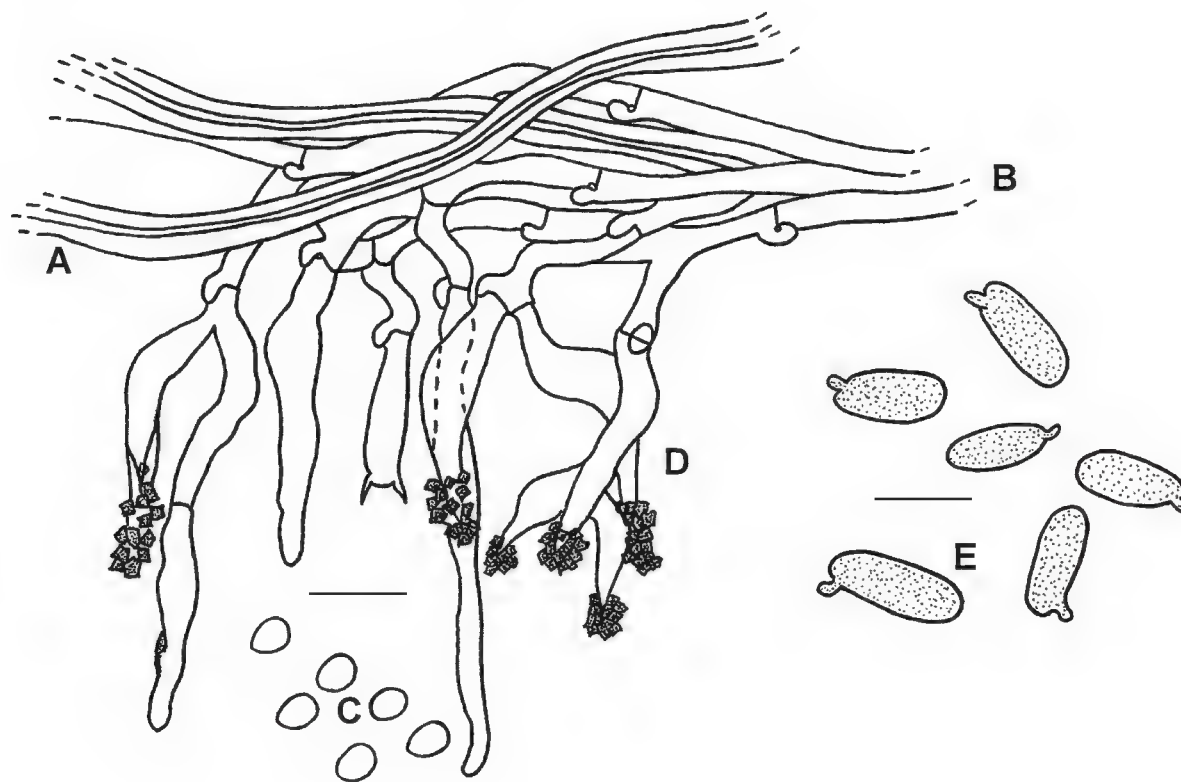


FIG. 6. *Echinoporia aculeifera*. Scale = 10  $\mu\text{m}$ .

A. Skeletal hyphae. B. Generative hyphae. C. Basidiospores.  
D. Cystidia. E. Conidiospores.

COMMENTS: The species is easily recognized in the field by the dense cover of long yellowish-orange to red hairs (hydroid processes) and irregular pores. *Echinoporia aculeifera* produces abundant conidiospores, absent in other polypores, as pointed out by Gilbertson & Ryvarden (1986). Wright (1983) reported rare cystidia with a crystal crown ( $11.3\text{--}21.7 \times 4.1\text{--}5.2 \mu\text{m}$ ). Our collection had abundant cystidia and incrustated hyphal terminations. The basidiospore size ( $5\text{--}7 \times 3\text{--}4 \mu\text{m}$ ) agrees with that cited by Silveira & Guerrero (1991). However, Gilbertson & Ryvarden (1986) noted smaller basidiospores ( $4\text{--}5 \times 3\text{--}3.5 \mu\text{m}$ ).

ADDITIONAL MATERIAL: ARGENTINA, Misiones, Cataratas del Iguazú, Singer & Digilio, M-132, 27/XI/49 (BAFC 27280); *ibid*, Parque Nacional Iguazú, plaza cerca Salto Dos Hermanos, J.E. Wright, M-3028, 28/IX/79 (BAFC 24462).

DISTRIBUTION: Neotropical; Brazil (Bahia, Rio Grande do Sul, Paraná and São Paulo), North American, Central America and South America (Fonsêca 1999, Gilbertson & Ryvarden 1986, Góes-Neto 1999, Popoff & Wright 1998, Rajchenberg & Meijer 1990, Silveira & Guerrero 1991).

*Oxyporus obducens* (Pers.) Donk, Med. Bot. Mus. Univ. Utrecht 9: 202, 1933. FIG. 7  
= *Polyporus obducens* Pers., Mycol. Eur. 2: 104, 1825.

DESCRIPTION: Núñez & Ryvarden (2001).

VOUCHER MATERIAL: BRAZIL, Santa Catarina: Mondaí, Linha Uruguai, Campos-Santana & Santana 213, 23/V/07 (FLOR 32223).

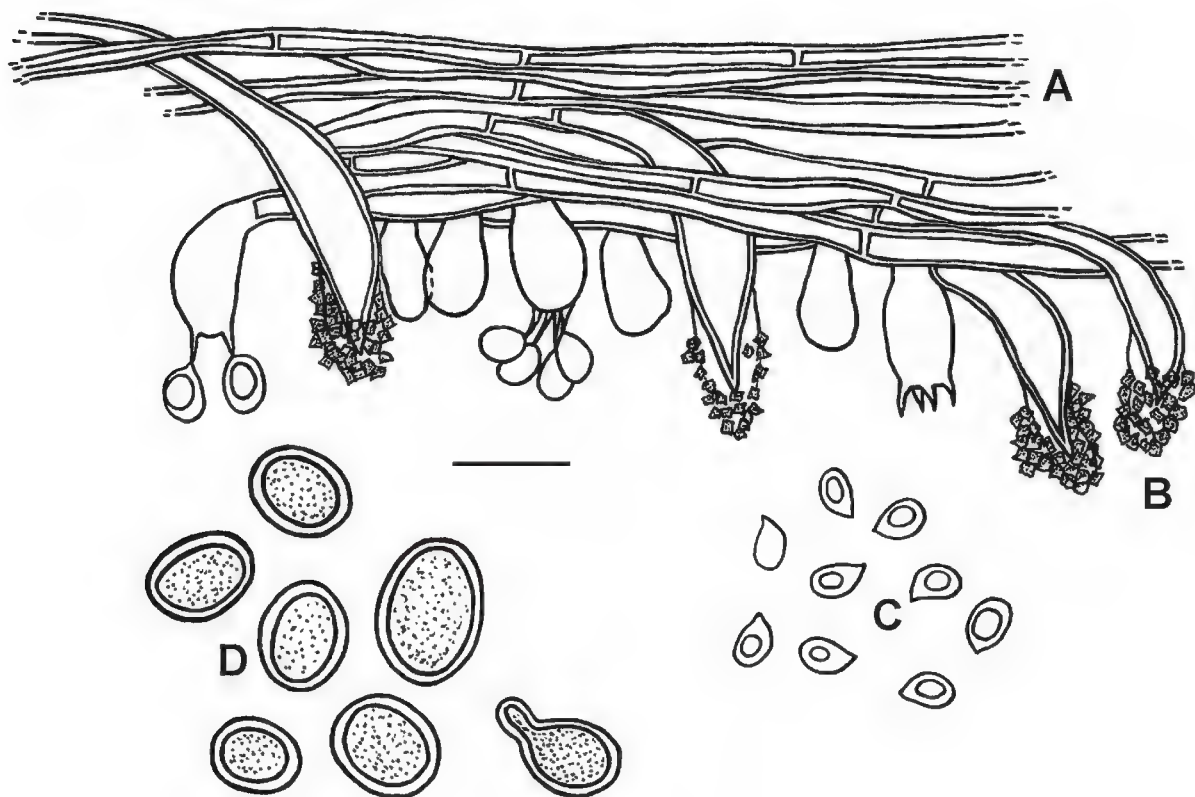


FIG. 7. *Oxyporus obducens*. Scale =  $10 \mu\text{m}$ .

A. Generative hyphae. B. Cystidia. C. Basidiospores. D. Chlamydospores.

COMMENTS: The specimen studied differs from other resupinate *Oxyporus* species by the number of pores (4–6 per mm), basidiospore size ( $3\text{--}5\text{--}(6) \times 3\text{--}4 \mu\text{m}$ ), and the presence of chlamydospores. There are few discrepancies between our observations and the literature. Our collection agrees with Núñez & Ryvarden (2001), who recorded similarly sized cystidia ( $25\text{--}55 \times 7\text{--}8 \mu\text{m}$ ) and basidiospores ( $4\text{--}5 \times 2.5\text{--}3.0 \mu\text{m}$ ). Ryvarden & Gilbertson (1994) reported slightly smaller basidiospores ( $3\text{--}4.5 \times 2.5\text{--}3.5 \mu\text{m}$ ) and cystidia ( $15\text{--}30 \times 5\text{--}12 \mu\text{m}$ ).

ADDITIONAL MATERIAL: BRAZIL, Santa Catarina: Santo Amaro da Imperatriz, Morro das Três Voltas, Michels, Esber, Groposo & Marcon-Baltazar 496, 20/III/2005 (FLOR 31806); *ibid*: Florianópolis, Ratones, Loguercio-Leite & Furlani 383, 27/I/1989 (FLOR 10702).

DISTRIBUTION: Cosmopolitan; Brazil [Rio Grande do Sul], Argentina, China, Czechoslovakia, Finland, Japan, Russia, USA, Venezuela (Núñez & Ryvarden 2001, Dai et al. 2004, Ryvarden & Gilbertson 1994, Robledo et al. 2006, Ryvarden & Iturriaga 2001, Rick 1960).

*Amauroderma sprucei* (Pat.) Torrend, Brotéria Bot. 18: 121, 1920.

FIG. 8

= *Ganoderma sprucei* Pat., Bull. Soc. Mycol. France 10: 75, 1894.

DESCRIPTION: Decock & Herrera Figueroa (2006).

VOUCHER MATERIAL: BRAZIL, Santa Catarina: Mondaí, Linha Uruguai, Campos-Santana, Santana & Rodrigues-Souza 190, 27/XII/06 (FLOR 32210).

COMMENTS: The globose to subglobose basidiospores ( $9\text{--}10 \times 7\text{--}8 \mu\text{m}$ ) of our collection are similar in size to those ( $8.5\text{--}10 \times 7\text{--}9 \mu\text{m}$ ,  $9\text{--}10 \times 7\text{--}8 \mu\text{m}$ ) seen in the additional material (URM 77450; URM 77451) as well as those reported by Ryvarden (2004) and Furtado (1981;  $8\text{--}10 \mu\text{m}$ ,  $(6\text{--})8\text{--}10 \mu\text{m}$  in diam]. Decock & Herrera Figueroa (2006) observed basidiospores measuring  $(6.5\text{--})7.5\text{--}9.8$  ( $\text{--}10.3$ )  $\times$   $(6.5\text{--})7\text{--}9$  ( $\text{--}9.5$ )  $\mu\text{m}$ . Although Ryvarden (2004) describes *A. sprucei* as producing globose basidiospores, the Mondaí material (FLOR32210), URM 77450, and URM 77451 showed globose to subglobose basidiospores, matching the shape reported by Decock & Herrera Figueroa (2006). *Amauroderma sprucei* differs from other *Amauroderma* species known from Santa Catarina — *A. schomburgkii* (Mont. & Berk.) Torrend, *A. omphalodes* (Berk.) Torrend, *A. intermedium* (Bres. & Pat.) Torrend, *A. brasiliense* (Singer) Ryvarden, *A. camerarium* (Berk.) J.S. Furtado — by its reddish yellow hymenophore and dextrinoid skeletal hyphae.

ADDITIONAL MATERIAL: BRAZIL, Sergipe: Itabaiana, Estação Ecológica Serra de Itabaiana, Gibertoni 44616, III/2002 (URM 77450); *ibid*, Gibertoni 44617, III/2002 (URM 77451); *ibid*, Santa Catarina: Santo Amaro da Imperatriz, Hotel Caldas da Imperatriz, Larissa T. Pereira, 31/III/2007 (FLOR 32197); *ibid*, Vargem Braço — PEST, Groposo 110, 28/III/2001 (FLOR 31323); *ibid*, Trilha da Cascata — PEST, Groposo 097, 05/I/2001 (FLOR 11902); *ibid*, Florianópolis, Rio Tavares, Furlani 274, 04/VII/1986 (FLOR 10460); *ibid*, Ilhota — Morro do Baú, Groposo, VII/2003 (FLOR 31344).



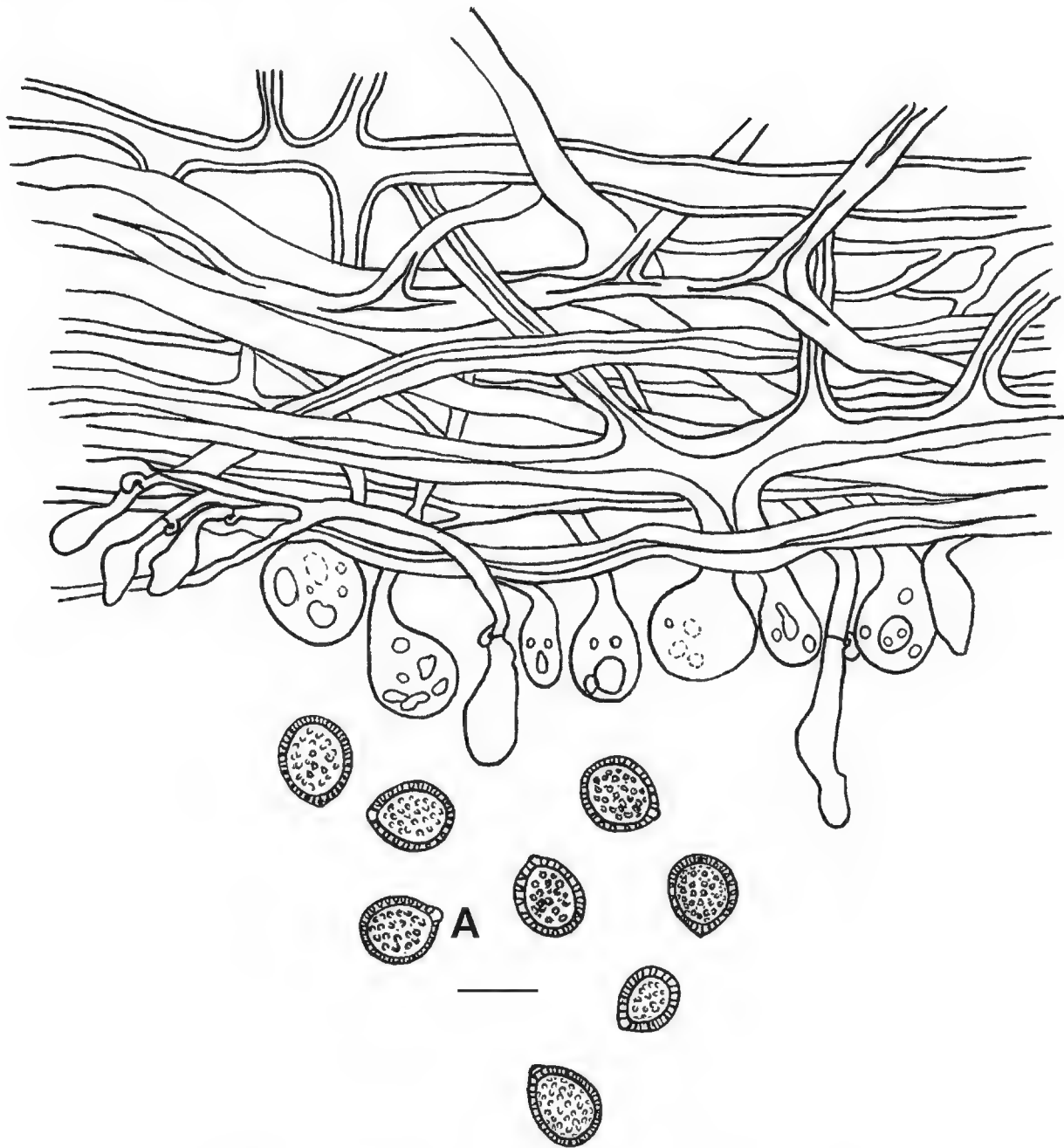


FIG. 8. *Amauroderma spruce* hymenium. Scale =10  $\mu$ m.  
A. Basidiospores.

DISTRIBUTION: Neotropical; Brazil (Amazonas, Rio Grande do Sul, Minas Gerais, Mato Grosso, Pernambuco, Rio de Janeiro, São Paulo, Paraná and Sergipe), Costa Rica, Cuba, Belize, French Guyana and Venezuela (Torrend 1920, Rick 1938, Furtado 1981, Ryvarden & Meijer 2002, Gibertoni 2004, Corner 1983, Ryvarden 2004, Decock & Herrera Figueroa 2006).

*Pseudofavolus miquelii* (Mont.) Pat., Essai Tax. Hymenomyc.: 81, 1900. FIG. 9  
= *Polyporus miquelii* Mont., Ann. Sci. Nat., Bot., 3e Sér., 4:357, 1845.

DESCRIPTION: Ryvarden & Johansen (1980).

VOUCHER MATERIAL: BRAZIL, Santa Catarina: Mondaí, Linha Sanga Forte, Campos-Santana, Santana & Zanella 109, 16/VI/06 (FLOR 32225).

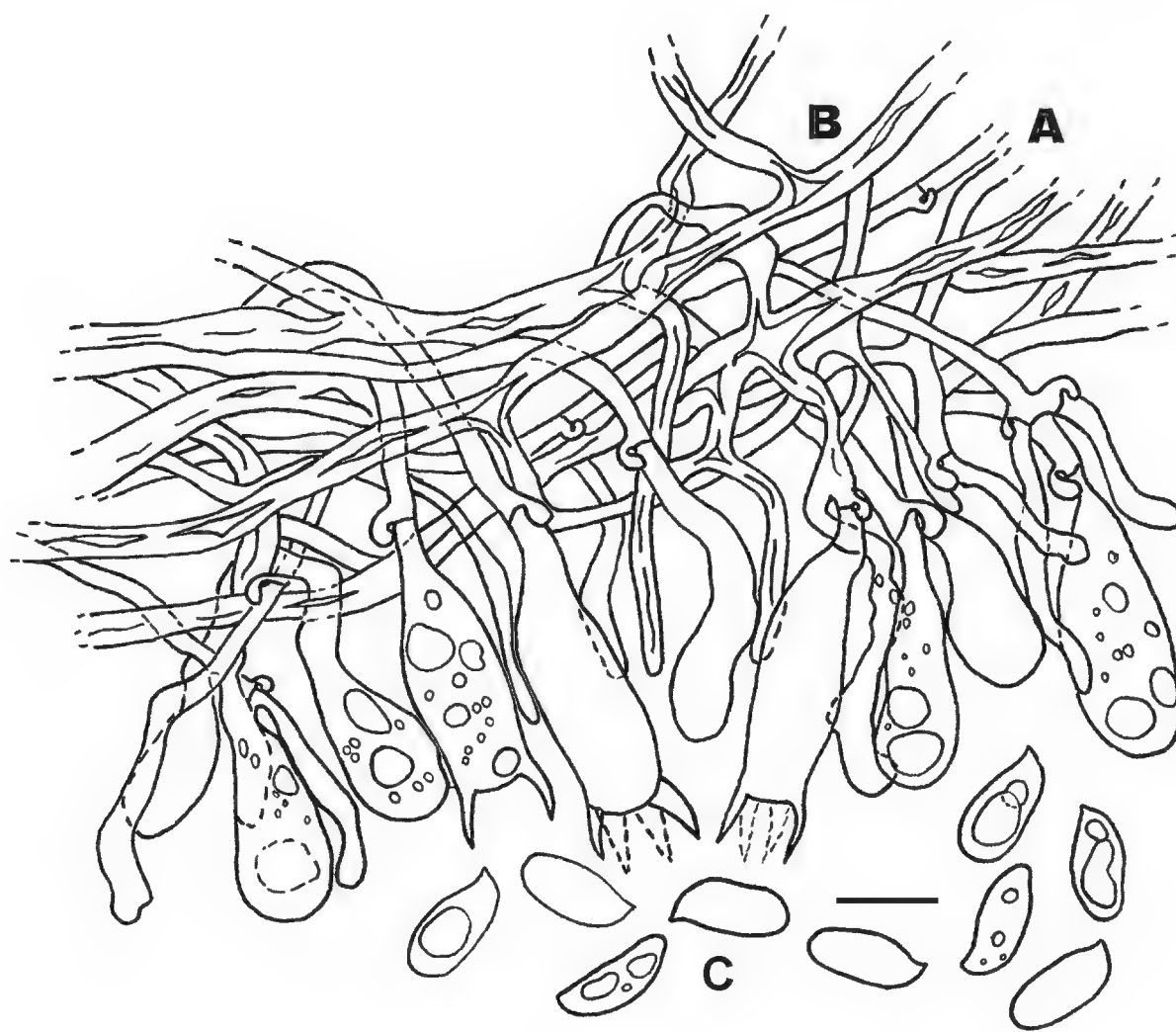


FIG. 9. *Pseudofavolus miquelii*. Scale = 10  $\mu$ m.  
A. Generative hyphae. B. Skeleto-binding hyphae. C. Basidiospores.

COMMENTS: Núñez & Ryvarden (1995) characterized *P. miquelii* as having a very thin context, large and angular pores, and basidiospores greater than 15  $\mu$ m long. Our basidiospores (10–16  $\times$  4–6  $\mu$ m) are very similar to material from Costa Rica (NYBG 00354169, NYBG 00354168: 10–17  $\times$  5–7  $\mu$ m) and slightly smaller than those recorded by Ryvarden & Johansen (1980; (14.5–)16–20  $\times$  6.5–8.0  $\mu$ m) and Corner (1984; 12–18  $\times$  6–8.5  $\mu$ m). Ryvarden & Johansen (1980) pointed out that the absence of a cuticle, the very thick context (1–2 mm), and number of the pores per mm ((1–)2–3) separate this species from *Pseudofavolus cucullatus* (Mont.) Pat. Corner (1984) considered *P. cucullatus* a variety of *Polyporus miquelii*.

ADDITIONAL MATERIAL: COSTA RICA, El Jardín, Dota, L. Echeverría 41–78, 21/III/1900 (NYBG 00354169); *ibid*, SJ Montana, L. Echeverría 65–78, /1900 (NYBG 00354168); BRAZIL, Santa Catarina: Santo Amaro da Imperatriz, Morro das Três Voltas, Michels, Esber, Groposo e Marcon-Baltazar 494, 20/III/2005 (FLOR 31805); *ibid*, Ilha de Santa Catarina, Rio Vermelho, Loguercio-Leite, 14/XII/1984 (FLOR 10104); *ibid*, Paraná, Capanema, Basso, 27/XII/1996 (FLOR 11500).

DISTRIBUTION: Pantropical; Brazil (Mato Grosso do Sul), Australia, Africa, Paraguay and Costa Rica (Ryvarden & Johansen 1980, Núñez & Ryvarden 1995, Popoff & Wright 1998, Velázquez & Ruíz-Boyer 2005).

### Acknowledgments

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## MYCOTAXON

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**A phylogenetic study of *Trechispora thelephora***STEVEN ALBEE-SCOTT<sup>1\*</sup> & BRADLEY R. KROPP<sup>2\*\*</sup>

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**Abstract** — Molecular data support the recent transfer of *Hydnodon thelephorus* to the genus *Trechispora*. These data also provide preliminary evidence that the pileate-stipitate basidiome morphology of *Trechispora thelephora* is ancestral to the resupinate morphology typical of the genus *Trechispora*. A photo, description, and line drawings of *Trechispora thelephora* are provided.

**Key Words** — *Basidiomycota*, nuclear large subunit, phylogeny

**Introduction**

*Trechispora thelephora* is a relatively common fungus that is widespread in the neotropics (Cifuentes et al. 2005, Ryvarden 2002). In spite of this and its rather striking morphology (FIG. 1), it has received relatively little attention from mycologists until recently. The basionym of *Trechispora thelephora* is *Hydnum thelephorum* (Léveillé 1844), but it was later placed in the monotypic genus *Hydnodon* (Banker 1913) where it remained for 89 years until Ryvarden (2002) proposed transferring it to the genus *Trechispora*.

Even though the micromorphology of *T. thelephora* corresponds very well to the genus *Trechispora*, the pileate-stipitate morphology of its basidiomata is unusual for this usually resupinate genus (FIG. 1a, b, c). Perhaps as a consequence of this, the nomenclatural history of *T. thelephora* is fairly complex. This has been reviewed by Cifuentes et al. (2005) and Ryvarden (2002), but molecular work would help understanding of the classification of this fungus. Our goals were to study the phylogenetics of *Trechispora thelephora*. We provide a description, photograph, and line drawings of this rarely illustrated taxon.

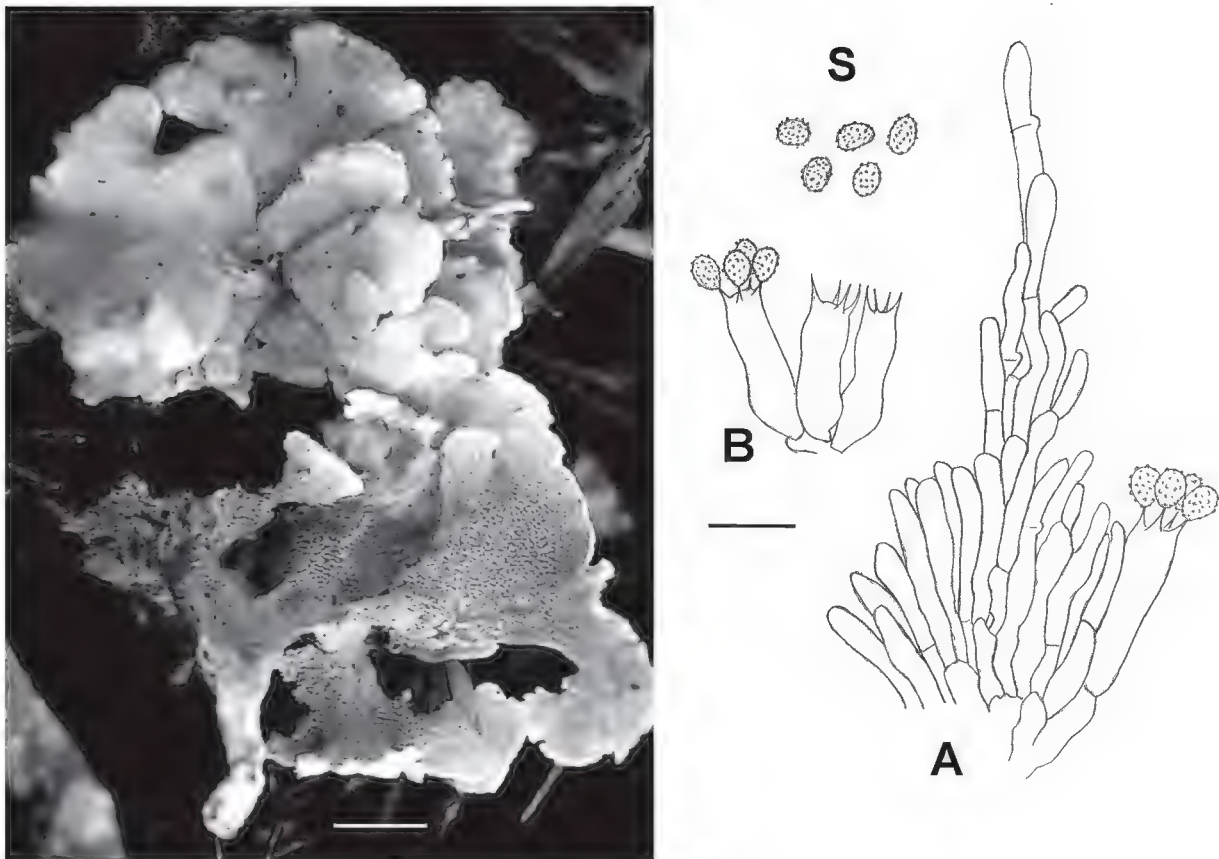


FIGURE 1. a) Pileate-stipitate basidiomata of *Trechispora thelephora* (UTC252606, Brad Kropp 13-Oct-02-23). Scale = 10 mm. b) Micromorphology of *Trechispora thelephora* showing section through aculeus (A), basidia (B), and basidiospores (S). Scale = 10  $\mu$ m.

### Material and methods

DNA was extracted from a basidiome of *T. thelephora* and sequence data was obtained for the portion of the nuclear large ribosomal subunit (nLSU) between primers LROR and LR5 (Moncalvo et al. 2000) and deposited in Genbank (HM104485). Sequences for additional taxa were downloaded from Genbank and aligned using ClustalX (Thompson et al. 1997). Taxon sampling included *Gloeocystidiellum porosum* (Berk. & M.A. Curtis) Donk, *Tubulicium vermiferum* (Bourdot) Jülich., and 12 members of the genus *Trechispora*. *Tubulicium vermiferum* was used as outgroup because it is sister to *Trechispora* according to Larsson et al. (2004). *Gloeocystidiellum porosum*, more distantly related to *Trechispora* (Larsson et al. 2004), was used to further polarize the crown group. A gap open of 5 and a gap extension of 1 for both pairwise and multiple alignment were used for the alignments. MrBayes 3.1 (Ronquist & Huelsenbeck 2003) was used to search tree space. All searches were performed using a time reversible model of evolution (Maddison 1994, Rodriguez et al. 1990) under the assumption of a discrete gamma distribution with six substitution types and some invariant sites (GTR+G+I). Posterior probabilities were approximated by sampling every hundred trees simulated using the Metropolis-coupled Markov chain Monte Carlo (MCMCMC) method. All runs were conducted with eight active MCMCMC chains, heated at 0.2, and started with a neighbor-joining tree to avoid entrapment in a local minimum. All runs were

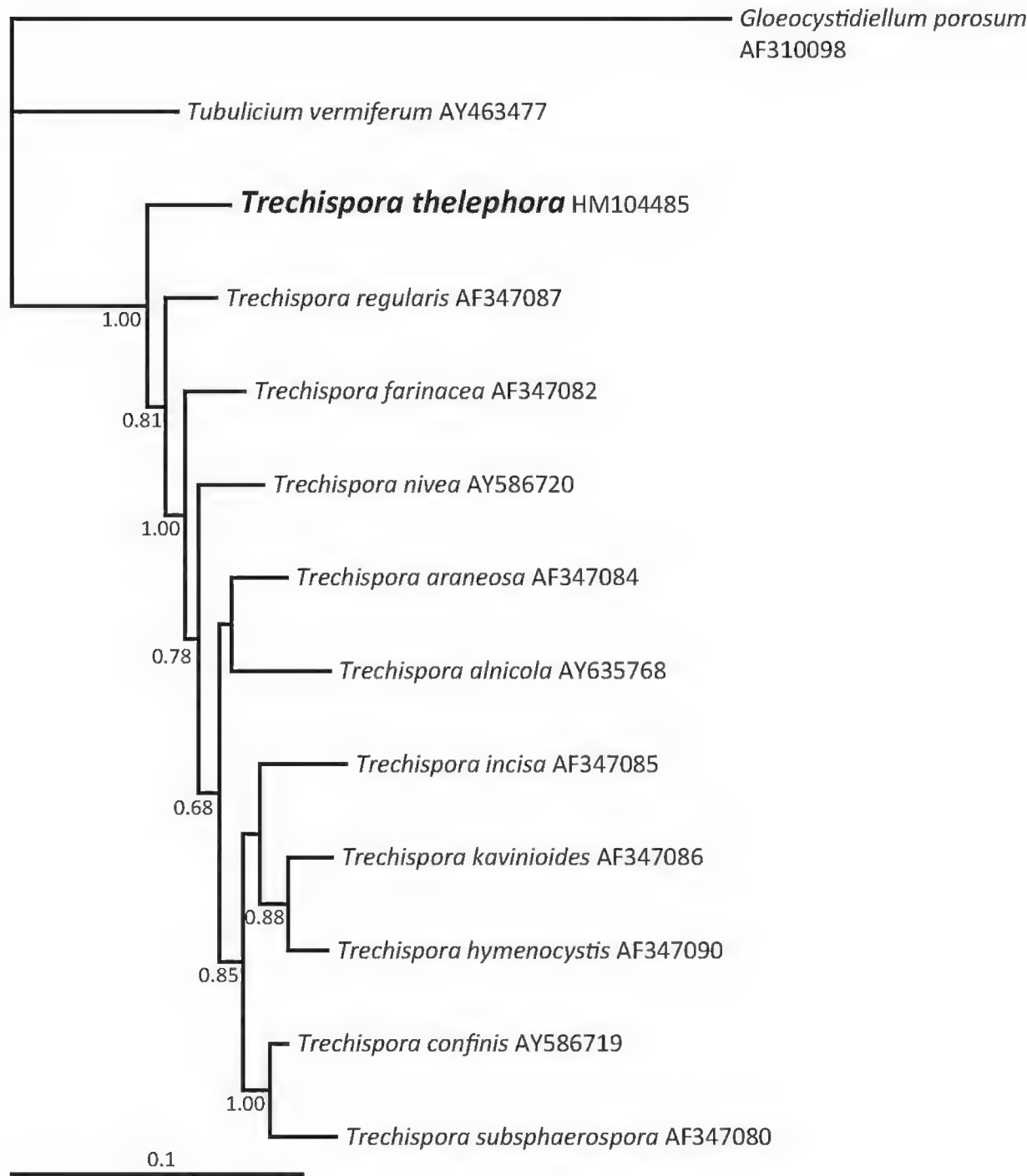


FIGURE 2. Phylogram derived from a Bayesian analysis of nLSU sequences from eleven *Trechispora* species. The phylogram has been rooted with *Gloeocystidiellum porosum*. Support measures are shown for nodes with posterior probability support of greater than 50 percent.

iterated for 1,000,000 generations. A majority consensus tree was calculated from the last 7000 trees from all runs to recover the posterior probabilities of the internal nodes using the sumt command in MrBayes. TreeView (Page 1996) was used to visualize the output from each simulation. Support measures for nodes with less than a 50% posterior probability support are not shown in FIG. 2.



Microscopical study and confirmation of the identity of our specimen was carried out using a light microscope after rehydrating sections in 10%  $\text{NH}_4\text{OH}$ . Microscopical measurements were done using oil immersion at 1000 $\times$  and line drawings were made with the aid of a drawing tube. The specimen from which DNA was extracted and from which the description and illustrations in FIG. 1 were made has been accessioned into the Intermountain Herbarium (UTC252606) at Utah State University.

## Results

A BLAST search using nLSU sequence data obtained from our specimen of *Trechispora thelephora* matched other *Trechispora* sequences, supporting the proposal (Ryvarden 2002) placing the species within *Trechispora*. Results of the phylogenetic analysis (FIG. 2) of nLSU sequences from other *Trechispora* taxa also support this and provide preliminary evidence that the pileate-stipitate basidiome morphology of *T. thelephora* is ancestral to the resupinate morphology that is typical for most of the genus.

Although basidiome morphology in the *Holobasidiomycota* can apparently evolve either toward or away from complex pileate-stipitate forms, Hibbett & Binder (2002) indicate that the rate of change from resupinate toward pileate-stipitate forms exceeds the rate of change away from pileate-stipitate forms. A later study by Hibbett (2004) also supports an overall evolutionary trend in the *Holobasidiomycota* toward pileate-stipitate basidiomata, indicating that the ancestral form in this group is probably resupinate, even though results vary depending on the analytical method used.

The analysis of our sequence data with additional data from Genbank allows us to postulate that within *Trechispora* evolution has favored simplification of basidiome morphology and that the predominantly resupinate *Trechispora* basidiomata have evolved from a pileate-stipitate ancestral state. However, further work, perhaps including another pileate-stipitate species, *Trechispora gillesii* (Maas Geest.) Liberta (Liberta 1973), should be done to support this observation.

*Trechispora thelephora* (Lév.) Ryvarden, Synopsis Fung. 15: 32 (2002) FIG. 1

Basidiome pileate-stipitate, upper surface light yellow brown, glabrous or with appressed fibrils, divided into multiple irregular lobes 24–12 mm across; lower hymenial surface pinkish, lighter toward margins, finely hydroid with teeth 1.0–0.5 mm in length, running part way down the stipe; hymenium drying soft with the subhymenial context drying hard and brittle. Stipe 5 mm wide  $\times$  20 mm tall, glabrous, concolorous with upper surface of basidiome or pallid near the base. Context pallid and not changing color when cut. Odor pleasant, fungoid. Spore print faint salmon. Hyphal system monomitic, hyphae of hymenial layer 2.0–3.5  $\mu\text{m}$  wide, thin walled, with clamps, hyphae

of subhymenial context dense, with clamps, slightly thick walled (walls up to 0.5 µm thick), 3.0–5.5 µm wide. Basidia clavate, with four sterigmata and basal clamps, 15–23 × 5–6 µm. Basidiospores ellipsoid, echinulate, 4.0–5.0 × 3.4–4.5 µm.

SPECIMEN EXAMINED — BELIZE. Cayo District, LAS CUEVAS RESEARCH STATION, Brad Kropp 13-Oct-02-23 (UTC 252606).

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## MYCOTAXON

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**A new species of *Podosporium*  
and a new record from southern China**

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**Abstract** — Two conidial fungi, *Podosporium cyclocaryae* sp. nov. and *Endophragmiella theobromae*, occurring on dead branches of *Cyclocarya paliurus* and *Dendrocalamus giganteus*, respectively, are described and illustrated and compared with related taxa. The specimens were collected from tropical forests in Fujian Province, China.

**Key words** — anamorphic fungi, taxonomy

**Introduction**

There is an enormous diversity of anamorphic fungi growing on rotten wood and dead branches in the tropical forests of southern China, and several mycological investigations dealing with many new species have been recently published (Yuan & Dai 2008, Zhang et al. 2009, Dai et al. 2009). Two additional species have been found that are described below. One is proposed herein as a new species and the other is a new record for China. The type specimen is deposited in HSAUP (Herbarium of the Department of Plant Pathology, Shandong Agricultural University) and HMAS (Mycological Herbarium, Institute of Microbiology, Chinese Academy of Sciences).

**Taxonomy*****Podosporium cyclocaryae* Y.D. Zhang & X.G. Zhang, sp. nov.**

FIG 1

MYCOBANK MB 518832

*Coloniae in substrato naturali effusae, brunneae. Mycelium hyalinum, hyphae ramosae, pallide brunneae, septata, 3–4 µm crassis. Conidiomata synnemata, solitaria, erecta, atrobrunnea vel nigra, cylindrica, usque 490 µm alta, 39–49 µm crassa ad basim, saepe inflata. Conidiophora macronematosa, synnematosae, nonramosae, septatae, laevia, brunnea*

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\*Corresponding author



FIG. 1. *Podosporium cyclocaryae*.  
A. Synnemata B. Conidiophores with conidia. C. Conidia.

*vel atrobrunnea, usque 490 µm longa, 3.5–4.5 µm crassa, divergentia ad apicem et lateralia. Cellulae conidiogenae monotretica, cylindrica, integratae, terminales, determinatae, laeves, brunneae vel atrobrunneae, 6.5–10 µm longae, 2.5–3.5 µm crassa. Conidia solitaria, sicca, acrogena, obclavata, stricta vel leviter curvatae, rostrata, 7–10-septata, laevia, 77–122 × 10–15 µm, brunnea, pallidiora versus apicem, basim cum depresso hilo, 2.5–3 µm crasso, praedita.*

HOLOTYPE: on dead branches of *Cyclocarya paliurus* (Batalin) Iljinsk. (Juglandaceae). Fujian Province, China, 11 Aug. 2009, Y.D. Zhang, HSAUP 0129 (isotype HMAS 146112).

ETYMOLOGY: in reference to the host genus, *Cyclocarya*.

Colonies on the natural substratum effuse, brown. Mycelium hyaline, hyphae flexuous branched, pale brown, septate, 3–4 µm thick. Conidiomata solitary, synnematos, erect, dark brown to black, cylindrical, scattered, up to 490 µm high, 39–49 µm wide at the often swollen base. Conidiophores macronematous, arranged in synnemata, unbranched, septate, smooth, brown to dark brown, up to 490 µm long, 3.5–4.5 µm wide, diverging laterally and also terminally. Conidiogenous cells monotretic, cylindrical, integrated, terminal, determinate,



smooth, brown to dark brown,  $6.5\text{--}10 \times 2.5\text{--}3.5\ \mu\text{m}$ . Conidia solitary, dry, acrogenous, obclavate, straight to slightly curved, rostrate, 7–10-septate, smooth-walled,  $77\text{--}122 \times 10\text{--}15\ \mu\text{m}$ , brown, paler toward the apex, base with a depressed hilum,  $2.5\text{--}3\ \mu\text{m}$  wide.

NOTES: The genus *Podosporium* was established by Schweinitz (1832), based on *P. rigidum* Schwein. After the holotype was discovered to be missing, Ellis (1971) lectotypified *P. rigidum* by specimens collected on dead stems and branches of *Ampelopsis* and *Rhus* from U.S.A. *Podosporium* is characterized by darkly pigmented and cylindrical synnemata consisting of distinct conidiophores terminating in monotretic, percurrent to rarely sympodial, clavate or cuneiform conidiogenous cells and brown, acrogenous, multiseptate, obclavate conidia (Ellis 1971, Chen & Tzean 1993). Worldwide, more than 60 species of *Podosporium* have been validly described. Only *P. elongatum* has been reported from China (Chen & Tzean 1993). Most species grow as saprobes on rotten wood and bark of various trees and shrubs or on dead herbaceous material. Of the known species, the conidia of *P. cyclocaryae* resemble those of *P. rigidum* (Schweinitz 1832) in having phragmoconidia. However, the conidia of *P. cyclocaryae* are rostrate and larger than those of *P. rigidum* ( $77\text{--}122 \times 10\text{--}15\ \mu\text{m}$  vs.  $40\text{--}70 \times 10\text{--}14\ \mu\text{m}$ ). In addition, the synnemata of *P. cyclocaryae* expand at the top and they are much shorter than those of *P. rigidum* (2 mm).

***Endophragmiella theobromae*** M.B. Ellis, More dematiaceous hyphomycetes.

144 (1976)

FIG 2

SPECIMENS EXAMINED: on dead branches of *Dendrocalamus giganteus* Munro (*Gramineae*), forest park of Wuyishan, Fujian Province, China, 18 Aug. 2009, Y.D. Zhang, HSAUP H3140 (duplicate HMAS 146113).

Colonies effuse, hairy, dark blackish brown to black. Mycelium in the substratum sparse, composed of septate, smooth, pale brown, branched hyphae  $2\text{--}3\ \mu\text{m}$  wide. Conidiophores macronematous, arising singly or sometimes fasciculate, branched, erect, straight or slightly flexuous, smooth, septate, brown, paler towards the apex, up to  $110\ \mu\text{m}$  long,  $7.5\text{--}8.5\ \mu\text{m}$  wide, sometimes swollen at the base, with 1–4 proliferations. Conidiogenous cells monoblastic, integrated, terminal, percurrent, cylindrical, tapered to a truncate apex. Conidial secession rhexolytic. Conidia obovoid to pyriform, usually 2-septate, basal cell pale brown, central cells and apical cell dark brown, smooth,  $17.5\text{--}30\ \mu\text{m}$  long,  $8.5\text{--}13\ \mu\text{m}$  wide, with a distinct basal frill derived from the distal end of the conidiogenous cell.

NOTES: The genus *Endophragmiella* B. Sutton was proposed and originally described by Sutton (1973) for two species: the type species *E. pallescens* B. Sutton and *E. canadensis* (Ellis & Everh.) B. Sutton. The genus is characterized

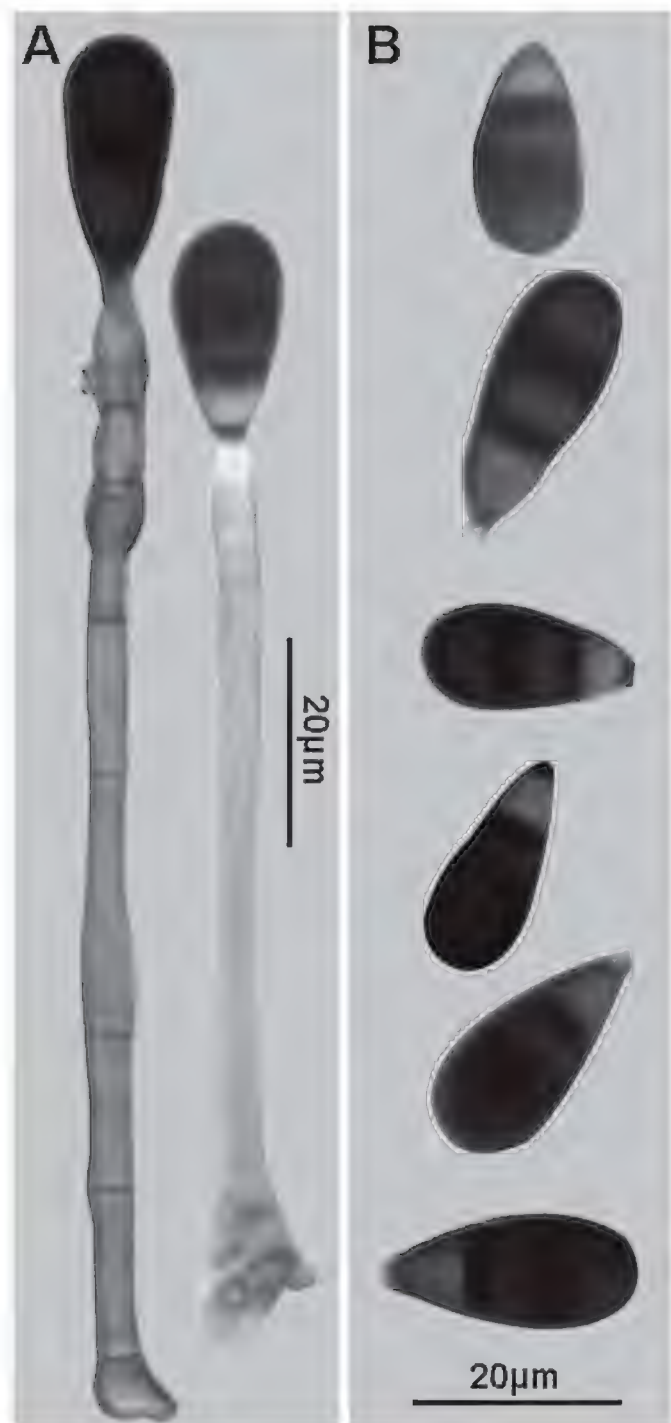


FIG. 2. *Endophragmiella theobromae*.  
A. Conidiophores with conidia. B. Conidia.

by conidiophores that are macronematous, mononematous with conidiogenous cells integrated, percurrent proliferation, and solitary, euseptate or distoseptate conidia with rhexolytic secession. The genus has been revised by Hughes (1979) and enlarged by Kirk (1985) and Holubová-Jechová (1986). At present, the genus *Endophragmiella* comprises more than 80 species, most of which grow as saprobes on rotten wood and bark of various trees and shrubs or on dead herbaceous material.

*E. theobromae* was first described by Ellis (1976) from New Guinea on dead cortex of *Theobroma cacao*. Our species was collected from a monocotyledonous plant (family *Gramineae*) in Fujian, China, whereas *E. theobromae* is known only from a dicotyledonous tree (family *Sterculiaceae*) in New Guinea. Our specimen is much similar to the type material, but the conidia in our collection are slightly larger and the conidiophores are smaller. Despite these minor differences, we believe they are the same species in different regions. This is the first record of this species from China.

### Acknowledgments

The authors are grateful to Dr Eric H.C. McKenzie and Dr R.F. Castañeda Ruiz for serving as pre-submission reviewers and for their valuable comments and suggestions. This project was supported by the National Natural Science Foundation of China (No. 30770015, 30499340, 2006FY120100).

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# MYCOTAXON

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## A new species of *Corynesporopsis* from Portugal

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**Abstract** — *Corynesporopsis iberica* sp. nov. found on the bark of an unidentified plant in Braganza, Portugal, is described and illustrated. It is characterized by an endogenous conidial ontogeny at the reduced internal area of inflated or globose bases of conidiophores, vase-shaped conidiogenous cells, and clavate to sub-cylindrical, (5–)7-septate, brown conidia with truncate bases and rounded apices. A key and illustrations to *Corynesporopsis* species is presented.

**Key words** — systematics, anamorphic fungi

## Introduction

Kirk (1981) erected the genus *Corynesporopsis* for a taxon previously placed in *Corynespora* Güssow, *Corynesporopsis quercicola* (Borowska) P. M. Kirk



(type species). The author remarked as primary characteristics of the genus *Corynesporopsis* the terminal, determinate or rarely with enteroblastic percurrent proliferations, monotretic conidiogenous cells and cylindrical to ellipsoid, euseptate, catenate conidia. Subsequently, eight other species have been added to this genus: *Corynesporopsis antillana* R.F. Castañeda & W.B. Kendr., *C. biseptata* (M.B. Ellis) Morgan-Jones, *C. cylindrica* B. Sutton, *C. inaequiseptata* Matsush., *C. indica* P.M. Kirk, *C. isabelicae* Hol.-Jech., *C. rionensis* Hol.-Jech., and *C. uniseptata* P.M. Kirk. Kirk (1981), Morgan-Jones (1988), Siboe & Kirk (1999), Castañeda et al. (2004), Siqueira et al. (2008), and McKenzie (2010) have noted that the distoseptate, solitary or catenate conidia that are borne through a slightly depressed and evident apical pore of the monotretic conidiogenous cell are distinctive characters of *Corynespora cassicola* (Berk. & M.A. Curtis) C.T. Wei (the most common species of *Corynespora*). Curiously, during direct isolation of *C. cassicola* from common leaf lesions on several hosts (*Cucumis sativus* L., *Solanum lycopersicum* L., *Vigna unguiculata* (L.) Walp., and others) only solitary conidia have been observed when the samples are examined directly from the field, but in pure cultures or after incubation in damp chambers, mostly catenate conidia with several enteroblastic cylindrical to doliiform percurrent proliferations of the conidiogenous cells can be observed. In fact, *C. cassicola* is a variable species that has been described several times as “new” based on small conidial size differences found on specimens collected on different hosts (Morgan-Jones 1988). However, these criteria are not sufficient to warrant recognition as novel species and the names should be reduced to synonyms of *C. cassicola* (Morgan-Jones 1988). Four other genera — *Briansuttonia*, *Corynesporina*, *Hemicorynespora*, and *Solicorynespora* — that are closely related to *Corynesporopsis* and *Corynespora* based on conidium ontogeny development (monotretic, determinate or sometimes doliiform to percurrent) can be separated by conidial production (solitary, basocatenate, or blastocatenate) and type of septa as circumscribing characters as summarized by Siqueira et al. (2008). During a November 2007 “Flora Micológica Ibérica” survey of microfungi in the Montesinho and Douro Natural Park, Braganza, Portugal, a conspicuous fungus from the genus *Corynesporopsis* was collected. The specimen showed differences from previously described taxa.

### Materials and methods

Samples of plant material were collected during a mycological survey in the Montesinho Natural Park, Braganza, Portugal. Individual collections were placed in paper and plastic bags taken to the laboratory and treated according to Castañeda (2005) and Castañeda et al. (2010). Mounts were prepared in polyvinyl alcohol-glycerol (8 g in 100 ml of water, plus 5 ml of glycerol) and measurements made at a magnification of  $\times 1000$ . Micrographs were obtained with a Zeiss Axioskop 40, Leitz Dialux 20 and a Jeol

JSM-6400 scanning electron microscope using the techniques described previously by Figueras & Guarro (1988).



FIG. 1. *Corynesporopsis iberica*, drawings from holotype (IMI 398785). Conidiophores, conidiogenous cells, and conidia. Scale bar = 10 µm.

## Taxonomy

*Corynesporopsis iberica* R.F. Castañeda, Silvera, Gené & Guarro, sp. nov.

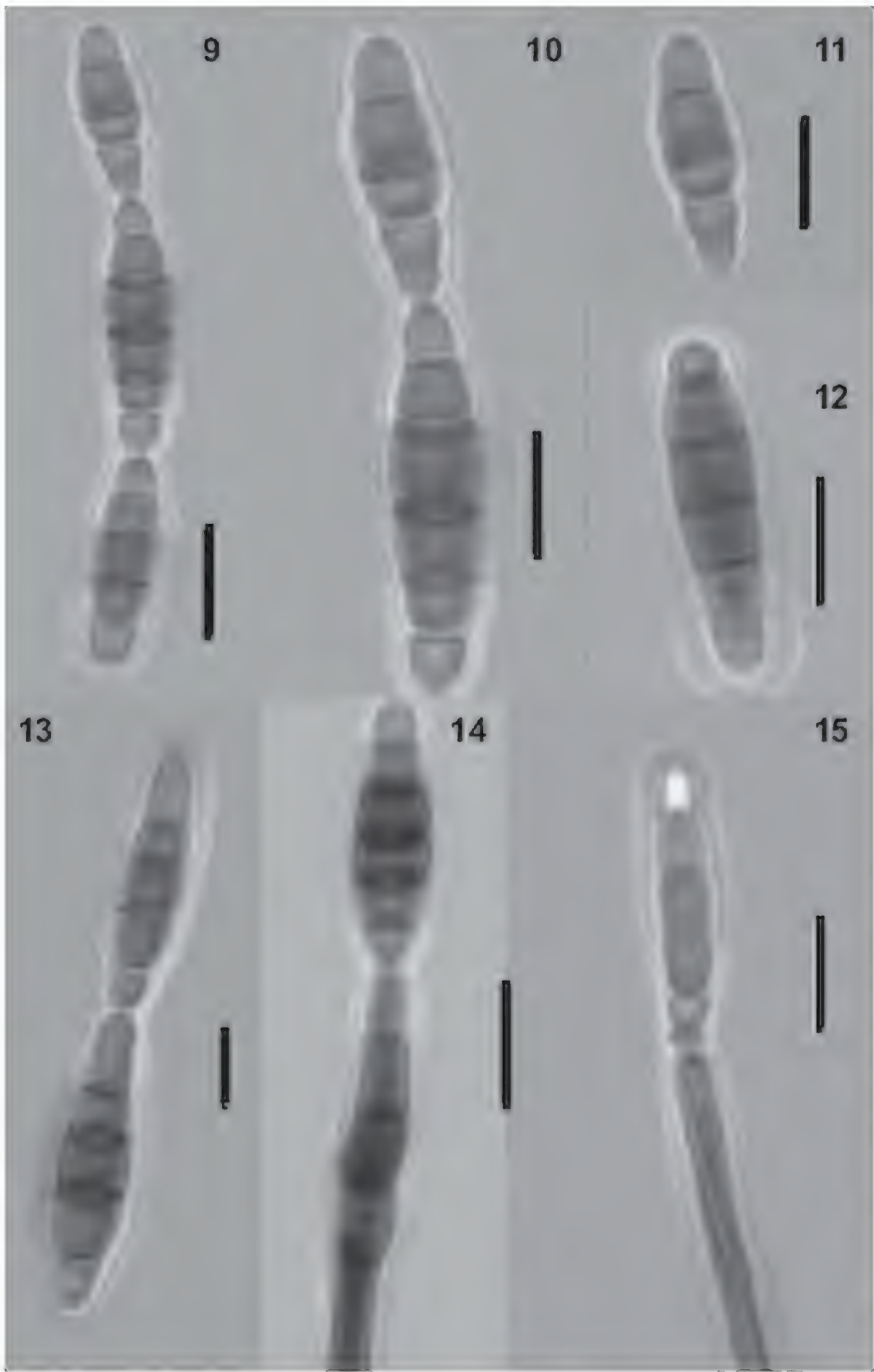
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FIGS 1–8

COLONIAE in substrato naturali effusae, pilosae, atrobrunneae vel nigrae. Mycelium plerumque in substrato immersum, ex hyphis septatis, cylindricis, aliquando cum cellulis inflatis, 1.5–2.5 µm diam., laevibus, atrobrunneis, compositum. CONIDIOPHORA



FIGS. 2–8. *Corynesporopsis iberica*, photomicrographs from holotype (IMI 398785).  
2. Conidia. 3–4. Conidiophores and conidiogenous cells. 5–8. photomicrographs (SEM) from culture derived from holotype. Conidiogenous cells and conidiogenous loci.  
Scale bars (1–4 = 10  $\mu\text{m}$ ; 5–8 = 3  $\mu\text{m}$ ).



FIGS. 9–15. *Corynesporopsis antillana*, photomicrographs from holotype (INIFAT C89/183).  
9–13. Conidia. 14–15. Conidiophores and conidiogenous cells. Scale bars = 10  $\mu$ m.

*mononematosa, macronematosa, simplicia, erecta, recta, cylindrica, 4–7-septata, laevia, atrobrunnea, 30–100 × 6–10 µm. CELLULAE CONIDIOGENAE monotreticae, terminal, determinatae, brunneae, 5–10 × 3.5–5.0 µm, cum parietibus incrassantis circa loco conidiogeno, praeditae. CONIDIA, cylindrica interdum leviter curvata, plus minusve utrimque rotundata, (2–)3–7-septata, atrobrunnea, laevia, sicca, 15–48(–59) × 3–4 µm, laevia, blastocatenulata. Teleomorphosis ignota.*

TYPE: PORTUGAL. BRAGANZA, MONTESINHO NATURAL PARK, on bark of an unidentified plant, 14.XI.2007. R.F. Castañeda, C. Silvera & J. Capilla (Holotype: IMI 398785; ex-type culture: FMR 9651, CBS).

ETYMOLOGY: Latin, iberica, in reference to Iberian Peninsula.

COLONIES on the natural substrate effuse, hairy, dark brown to black. Mycelium immersed; hyphae septate, branched, cylindrical and sometimes inflated, thickened cells, 1.5–2.5 µm diam., smooth-walled, dark brown. CONIDIOPHORES mononematous, macronematous, simple, erect, straight, cylindrical, 4–7-septate, smooth and thick-walled, 30–100 × 6–10 µm, dark brown. CONIDIOGENOUS CELLS monotretic, terminal, determinate, brown, 5–10 × 3.5–5.0 µm, markedly thick-walled around the conidiogenous loci. CONIDIA cylindrical, straight, sometimes slightly curved, more or less rounded at the ends, (2–)3–7-septate, with septa thick, smooth-walled, dark brown, 15–48(–59) × 3–4 µm, forming dark brown to black, acropetal, unbranched chains. Teleomorph unknown.

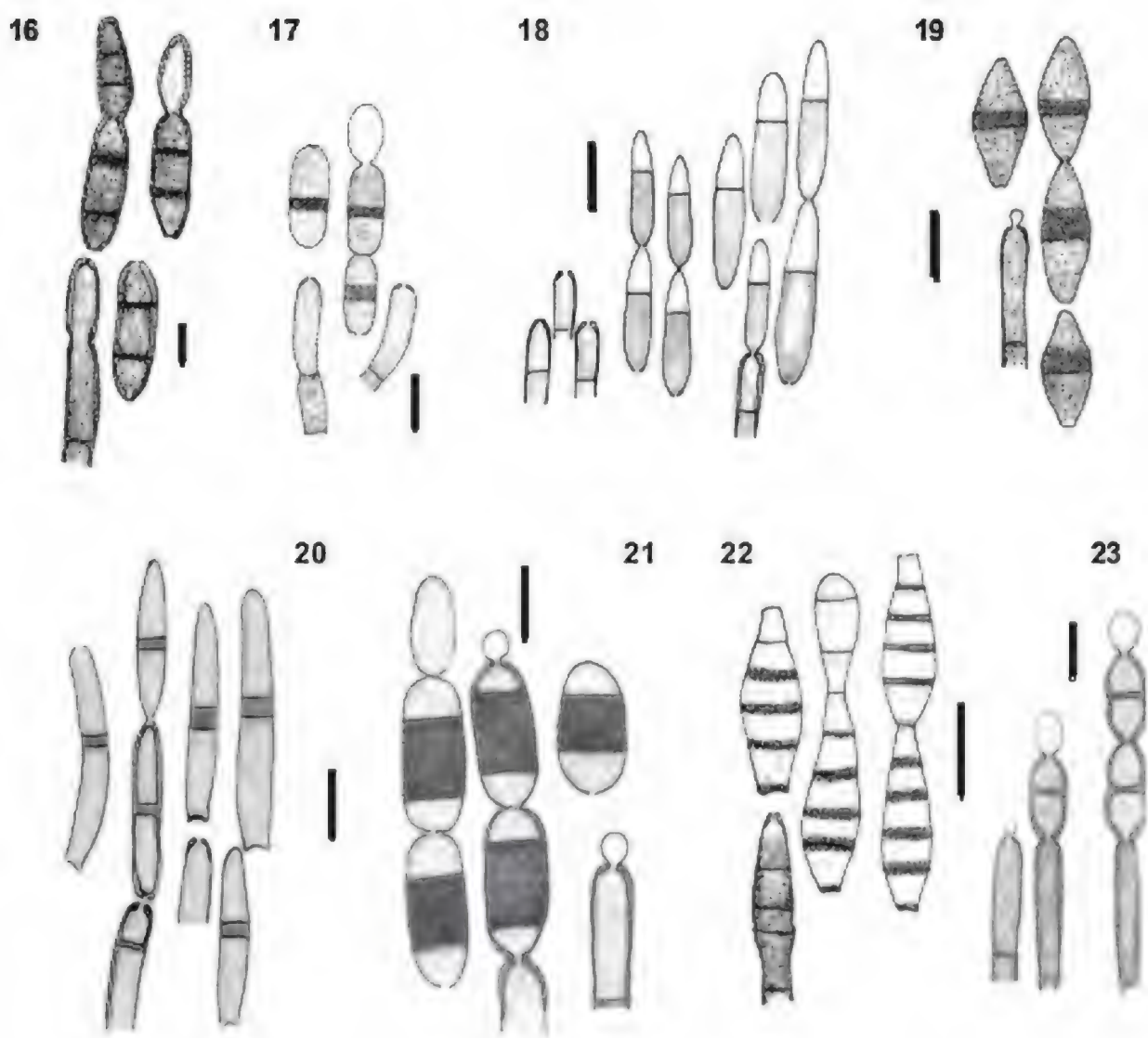
Culture from the holotype: COLONIES on corn meal agar mixed 1:1 with carrot extract, attaining 20–29 mm after 10 days at 25°C, floccose, pale brown. Reverse brown or cream-olivaceous. Hyphae thick-walled, septate, brown, 2–3 µm diam, smooth-walled. CONIDIOPHORES macronematous, cylindrical, multiseptate, smooth, brown, 3–8-septate, up to 160 µm tall, 5–8 µm wide. CONIDIA cylindrical, (2–)4–6-septate, dark brown to brown, smooth-walled, 15–48 × 3–4 µm, dry, blastocatenulate.

*Corynesporopsis iberica* slightly resembles *C. cylindrica*, but that species is easily differentiated by its shorter cylindrical conidiophores and brown, 1–2-septate, cylindrical, smooth, 12.5–20.5 × 6–7.5 µm conidia. Two other species with 3–5-septate conidia, *C. antillana* and *C. rionensis*, differ from *C. iberica* in shape and pigmentation.

**Key to *Corynesporopsis* species**

- 1    Conidia 1-septate ..... 2
- Conidia 1-septate, rarely 2-septate, cylindrical, smooth, medium brown,  
      guttulate, 12.5–20.5 × 6.0–7.5 µm ..... (FIG. 17) *C. cylindrica*
- Conidia with more than 1 septa ..... 3
- 2(1) Conidia elongate fusiform or navicular, smooth, brown, with the septum  
      dark and thick, 24–43 × 4–6 µm ..... (FIG. 20) *C. isabelicae*





FIGS. 16–23. *Corynesporopsis* spp., conidiogenous cells and conidia redrawn from the original descriptions. 16. *C. biseptata*. 17. *C. cylindrica*. 18. *C. inaequiseptata*. 19. *C. indica*. 20. *C. isabelicae*. 21. *C. quercicola*. 22. *C. rionensis*. 23. *C. uniseptata*. Scale bars = 10  $\mu$ m.

- Conidia ellipsoid to broadly obovoid, sometimes somewhat biconic, smooth,  
dark brown to very dark brown, with the septum obscured by a dark band,  
14–27  $\times$  8–14  $\mu$ m ..... (FIG. 19) *C. indica*
- Conidia broadly ellipsoid, manifestly constricted at the septum, smooth, brown, often  
darker at the septum, 12–16  $\times$  5–7  $\mu$ m ..... (FIG. 23) *C. uniseptata*
- Conidia narrowly obclavate, smooth, with brown basal cell and very pale brown  
apical cell, inequilateral, 17–25  $\times$  4.0–5.5  $\mu$ m..... ... (FIG. 18) *C. inaequiseptata*
- 3(1) Conidia usually 2-septate ..... 4
- Conidia usually with more than 2 septa. .... .5
- 4(3) Conidia broadly ellipsoid to cylindrical, smooth, end cells pale brown,  
middle cell dark brown, 12–22  $\times$  6–9  $\mu$ m ..... (FIG. 21) *C. quercicola*
- Conidia cylindrical, straight or slightly curved, smooth, pale to mid-brown,  
with central cell usually slightly longer than end cells,  
18–33  $\times$  7–9  $\mu$ m ..... (FIG. 16) *C. biseptata*

- 5(3) Conidia fusiform, broad fusiform or ellipsoidal, 3–4(–5)-septate, with septa dark and thick, distinctively truncate at the ends, smooth, brown or dark brown, apical cell pale brown or paler and apical cell of terminal conidium obtuse, 24–36 × 8–11 µm ..... (FIG. 22) *C. rionensis*
- Conidia broadly ellipsoidal to navicular, (3–)5(–6)-septate, constricted at the septa, slightly truncate or rounded at the ends, smooth, 3–4 central cells dark brown, septa black, pale brown or colorless at the ends, 21–33 × 5–8 µm ..... (FIGS. 9–15) *C. antillana*
- Conidia cylindrical, straight, sometimes slightly curved, (2–)3–7-septate, with the septa thick, rounded at the ends, smooth, dark brown, 15–48(–59) × 3–4 µm ..... (FIGS. 1–8) *C. iberica*

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**Taxonomic studies of *Ellisembia* from Hainan, China**

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**Abstract** — Two new species of the anamorphic genus *Ellisembia* were collected from tropical forests in Hainan Province, China. *Ellisembia podocarpi* sp. nov. and *E. photiniae* sp. nov., occurring respectively on dead branches of *Podocarpus imbricatus* and *Photinia parvifolia*, are described and illustrated. They are compared with similar species.

**Key words** — anamorphic fungi, taxonomy

**Introduction**

The genus *Ellisembia* was introduced by Subramanian (1992) to accommodate *Sporidesmium*-like species that have determinate or irregularly percurrently extending conidiogenous cells that produce distoseptate conidia. Wu & Zhuang (2005) merged *Imicles* Shoemaker & Hambl. (Shoemaker & Hambleton 2001) into *Ellisembia*, and expanded the generic concept to include typically lageniform, ovoid or doliiform percurrently extending conidiogenous cells. Following the generic concept of Subramanian (1992) and Wu & Zhuang (2005), more than 40 species have been described under *Ellisembia*, most of which are saprobes on rotten wood and dead branches of various plants (Subramanian 1992, McKenzie 1995, 2010, Goh & Hyde 1999, Mena & Delgado 2000, Zhou & Hyde 2001, Wu & Zhuang 2005, Heuchert & Braun 2006, Ma et al. 2008).

The tropical forests of Hainan have a rich mycota, and many wood-inhabiting fungi have been discovered there (Dai & Cui 2006, Zhang et al. 2009, Dai & Li 2010). During an ongoing mycological survey in these forests, numerous conidial fungi were collected on dead branches. Among these were two species having the morphological characteristics of genus *Ellisembia*. They differ

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significantly from previously described *Ellisembia* species and are therefore proposed as new taxa.

### Taxonomy

*Ellisembia podocarpi* Jian Ma & X.G. Zhang, sp. nov.

FIGS. 1–4

MYCOBANK MB 518837

*Fungus anamorphicus*. COLONIAE in substrato naturali effusae, brunneae, pilosae. Mycelium partim superficiale, partim immersum in substrato, ex hyphis ramosis, septatis, pallide brunneis, laevibus, 1.5–3 µm crassis compositum. CONIDIOPHORA macronemata, mononematica, singula vel fasciculata, erecta, nonramosa, recta vel flexuosa, cylindrica, brunnea, laevia, septata, 32–65 × 3–5.5 µm. CELLULAE CONIDIOGENAE monoblasticae, integratae, terminales, lageniformes vel cylindricae, brunneae, laeves, 8–16 × 3–4.5 µm, ad usque 0–3 proliferationes lageniformes vel doliiformes percurrentes. Conidiorum secessio schizolytica. CONIDIA holoblastica, solitaria, acrogena, recta vel curvata, obclavata, ad longa rostrata, laevia, brunnea vel pallide brunnea, 13–19-distoseptata, 110–170 µm longa (rostro incluso), 7.5–10 µm crassa, basi truncata 2–4 µm lata, cellula apicali versus attenuate, pallide brunnea vel subhyalina, aseptata, laevia, rostro, ad usque 80 µm longo, 1–2.5 µm lato.

HOLOTYPE: on dead branches of *Podocarpus imbricatus* Blume (*Podocarpaceae*), tropical forest of Jianfengling, Hainan Province, China. 3 May 2007, J. Ma, HSAUP H5281 (isotype HMAS 146080).

ETYMOLOGY: in reference to the host genus, *Podocarpus*.

Anamorphic fungi. COLONIES on natural substrate effuse, brown, hairy. Mycelium partly superficial, partly immersed in the substratum, composed of branched, septate, pale brown, smooth-walled hyphae, 1.5–3 µm thick. CONIDIOPHORES macronematous, mononematous, singly or in groups, erect, unbranched, straight or flexuous, cylindrical, brown, smooth, septate, 32–65 × 3–5.5 µm. CONIDIOGENOUS CELLS monoblastic, integrated, terminal, lageniform or cylindrical, brown, smooth, 8–16 × 3–4.5 µm, with 0–3 lageniform or doliiform percurrent proliferations. Conidial secession schizolytic. CONIDIA holoblastic, solitary, acrogenous, straight or curved, obclavate to long-rostrate, smooth-walled, brown to pale brown, 13–19-distoseptate, 110–170 µm long (rostrum included), 7.5–10 µm thick in the broadest part, 2–4 µm wide at the truncate base, apex extended into a pale brown to subhyaline, aseptate, smooth, rostrum, up to 80 µm long, 1–2.5 µm wide.

*Ellisembia podocarpi* is morphologically most similar to *E. filia* W.P. Wu (Wu & Zhuang 2005) and *E. maungatautari* McKenzie (McKenzie 2010), but differs from *E. filia* (conidia 40–50 µm long, 7–9-distoseptate) in having longer conidia with more numerous distosepta, and from *E. maungatautari* (conidia 13–15 µm wide, 17–23-distoseptate) in having narrower conidia with fewer distosepta. In addition, conidiophores of *E. podocarpi* extend percurrently up to 3 times while *E. filia* and *E. maungatautari* conidiophores do not extend.



FIGS. 1–4. *Ellisembia podocarpi*. 1, 2. Conidiophores with terminal conidia. 3, 4. Conidia.

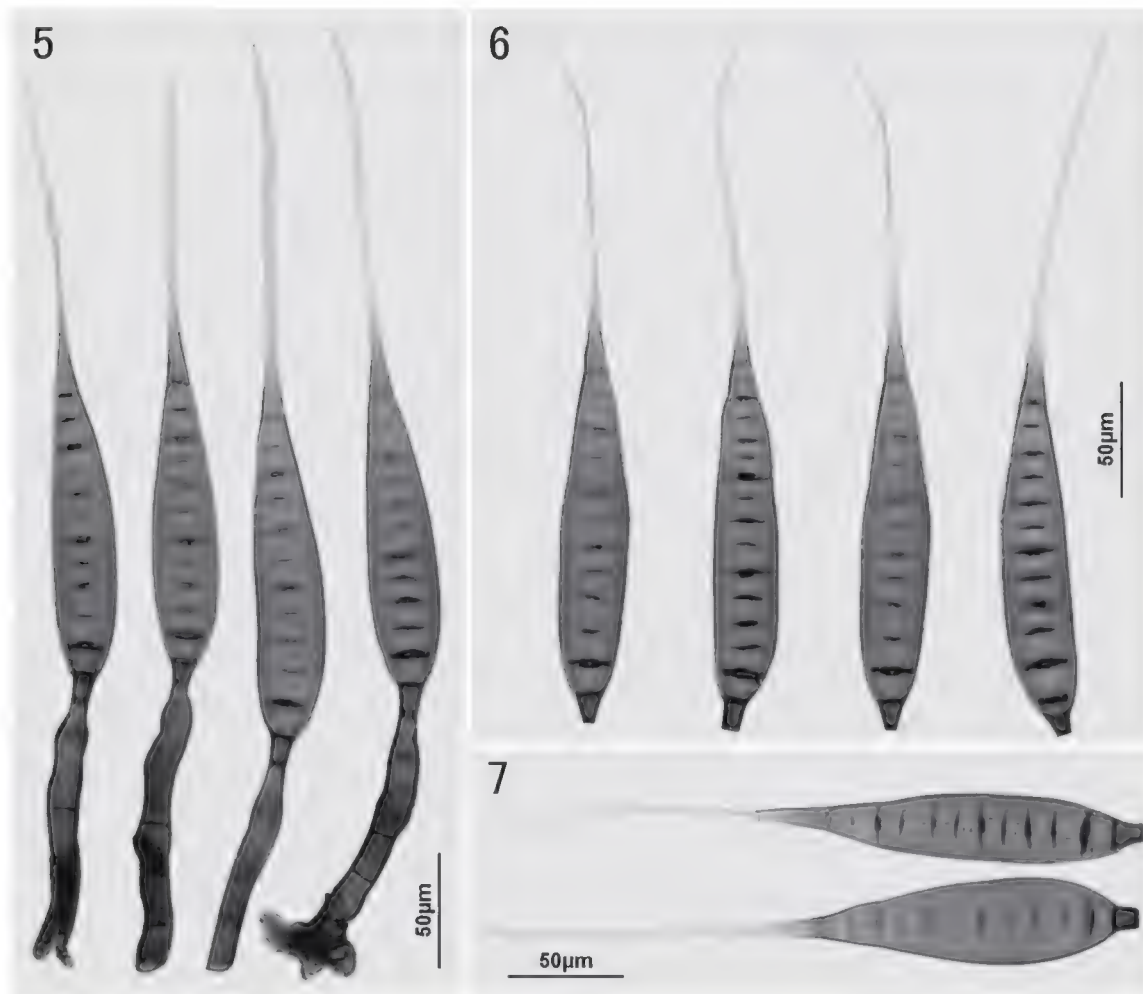
***Ellisembia photinia* Jian Ma & X.G. Zhang, sp. nov.**

FIGS. 5–7

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*Fungus anamorphicus*. COLONIAE in substrato naturali effusae, brunneae, pilosae. Mycelium partim superficiale, partim immersum in substrato, ex hyphis ramosis, septatis, pallide brunneis, laevibus, 1.5–2.5 µm crassis compositum. CONIDIOPHORA macronemata, mononematica, singula vel fasciculata, erecta, nonramosa, recta vel flexuosa, cylindrica, brunnea vel atrobrunnea, laevia, septata, 8.5–32 × 5.5–7.5 µm. CELLULAE CONIDIOGENAE monoblasticae, integratae, terminales, lageniformes vel cylindricae, brunneae, laeves, 27–30 × 6.5–7.5 µm, ad usque 0–1 proliferationes cylindricae percurrentes. Conidiorum secessio schizolytica. CONIDIA holoblastica, solitaria, acrogena, recta vel leviter curvata, obclavata, ad longa rostrata, laevia, brunnea vel pallide brunnea, 10–16-distoseptata, 92–170 µm longa (rostro incluso), 13–16 µm crassa, basi truncata 3–5 µm lata, cellula apicali versus attenuate, pallide brunnea vel subhyalina, aseptata, laevia, rostro 43–90 × 1–1.5 µm.

HOLOTYPE: on dead branches of *Photinia parvifolia* C.K. Schneid. (Rosaceae), tropical forest of Bawangling, Hainan Province, China. 10 Dec 2009, J. Ma, HSAUP H5189–4 (isotype HMAS 146081).



FIGS. 5–7. *Ellisembia photiniae*. 5. Conidiophores with terminal conidia. 6, 7. Conidia.

ETYMOLOGY: in reference to the host genus, *Photinia*.

Anamorphic fungi. COLONIES on natural substrate effuse, brown, hairy. Mycelium partly superficial, partly immersed in the substratum, composed of branched, septate, pale brown, smooth-walled hyphae, 1.5–2.5 µm thick. CONIDIOPHORES macronematous, mononematous, singly or in groups, erect, unbranched, straight or flexuous, cylindrical, brown to dark brown, smooth, septate, 8.5–32 × 5.5–7.5 µm. CONIDIOGENOUS CELLS monoblastic, integrated, terminal, lageniform or cylindrical, brown, smooth, 27–30 × 6.5–7.5 µm wide, with 0–1 cylindrical percurrent proliferations. Conidial secession schizolytic. CONIDIA holoblastic, solitary, acrogenous, straight or slightly curved, obclavate to long-rostrate, smooth-walled, brown to pale brown, 10–16-distoseptate, 92–170 µm long (rostrum included), 13–16 µm thick in the broadest part, 3–5 µm wide at the truncate base, apex extended into a pale brown to subhyaline, aseptate, smooth, rostrum 43–90 × 1–1.5 µm.

*Ellisembia photiniae* bears some resemblances to *E. filia* (Wu & Zhuang 2005) and *E. maungatautari* (McKenzie 2010) in conidial shape. However, conidia of *E. photiniae* are distinctly larger than those of *E. filia* (conidia 40–50 × 7–8

µm), and shorter than those of *E. maungatautari* (conidia 85–125 µm long). In addition, conidia of *E. photiniae* have 10–16 distosepta, while those of *E. filia* and *E. maungatautari* have 7–9 and 17–23 distosepta, respectively.

### Acknowledgments

The authors express gratitude to Dr W.B. Kendrick and Dr R.F. Castañeda Ruíz for serving as pre-submission reviewers and for their valuable comments and suggestions. This project was supported by the National Natural Science Foundation of China (Nos. 30499340, 30770015) and the Ministry of Science and Technology of the People's Republic of China (Nos. 2006FY120100, 2006FY110500–5).

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## MYCOTAXON

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**New records of *Corynesporopsis* from China**

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**Abstract** — Three species of *Corynesporopsis* — *C. uniseptata*, *C. quercicola*, and *C. indica* — are recorded for the first time from China. They are described and illustrated from specimens collected on dead branches of unidentified plants. The specimens are deposited in Herbarium of Shandong Agricultural University, Plant Pathology (HSAUP) and Mycological Herbarium, Institute of Microbiology, Chinese Academy of Sciences (HMAS).

**Key words** — hyphomycetes, taxonomy

**Introduction**

Kirk (1981a) established the genus *Corynesporopsis* for the single species previously known as *Corynespora quercicola*. It is characterized by single, differentiated, sometimes percurrent, conidiophores and integrated, terminal, monotretic conidiogenous cells that produce short acropetal chains of ellipsoid to cylindrical euseptate conidia. These characters separate *Corynesporopsis* P.M. Kirk from other similar genera including *Corynespora* Güssow (Güssow 1906), *Corynesporella* Munjal & H.S. Gill (Munjal & Gill 1961), *Hemicorynespora* M.B. Ellis (Ellis 1972), and *Solicorynespora* R.F. Castañeda & W.B. Kendr. (Castañeda & Kendrick 1990). Nine species are currently included in this genus, of which *Corynesporopsis quercicola* and *C. biseptata* (M.B. Ellis) Morgan-Jones were transferred from *Corynespora* (Kirk 1981a,b, 1983, Holubová-Jechová & Mercado 1986, Holubová-Jechová 1987, Morgan-Jones 1988, Sutton 1989, Castañeda Ruíz & Kendrick 1990, Matsushima 1993). Only *Corynesporopsis isabelicae* Hol.-Jech. has previously been reported from China (Lu et al. 2000). Most species are reported to survive saprophytically on dead branches, twigs,

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\*Corresponding author

and decaying leaves of various plants. During a continuing survey of tropical microfungi from the forests of Hainan Province of southern China, three species of *Corynesporopsis* were collected on dead branches. They are introduced as new records for China.

Taxonomy

*Corynesporopsis uniseptata* P.M. Kirk, Trans. Br. Mycol. Soc. 77(3): 463 (1981)

FIG. 1

SPECIMEN EXAMINED: on dead branches of unidentified plant, tropical forest of Bawangling, Hainan Province, China. 12 Dec 2009, J. Ma, HSAUP H5137 (duplicate HMAS 146082).



FIG. 1. *Corynesporopsis uniseptata*. Conidiophores and conidia.

ANAMORPHIC FUNGI. Colonies effuse, blackish brown to black, hairy. Mycelium partly superficial, partly immersed in the substratum, composed of branched, septate, pale brown to brown, smooth-walled hyphae, 2–4.5  $\mu\text{m}$  wide. Conidiophores differentiated, arising single or in groups on the hyphae, erect, straight or flexuous, unbranched, brown, smooth, septate, up to 160  $\mu\text{m}$  long, 3.5–5  $\mu\text{m}$  wide. Conidiogenous cells monotretic, integrated, terminal, determinate, cylindrical, brown, smooth, 16–35  $\mu\text{m}$  long, 4.5–6  $\mu\text{m}$  wide. Conidia acrogenous, dry, in acropetal chains of up to 10, ellipsoid to cylindrical, 1-euseptate, constricted at the septum, smooth, brown, often with darker pigmentation at the septum, 14–21  $\mu\text{m}$  long, 6–8  $\mu\text{m}$  wide in the widest part.

NOTES: *Corynesporopsis uniseptata* is reported for the first time from China. Compared with the morphology of the type specimen described by Kirk (1981b), the conidia of our collection are longer (14–21  $\mu\text{m}$  vs. 12–16  $\mu\text{m}$ ) and the conidiophores are also longer (up to 160  $\mu\text{m}$  vs. 60–100  $\mu\text{m}$ ), but we believe they are basically the same species. *Corynesporopsis uniseptata* most closely resembles *C. cylindrica* B. Sutton (Sutton 1989) in conidial shape and size range but differs in having didymospores with a median constriction at the septum. Moreover, the conidia of *C. cylindrica* are guttulate while *C. uniseptata* conidia are not.

***Corynesporopsis quercicola*** (Borowska) P.M. Kirk, Trans. Br. Mycol. Soc. 77(2):284 (1981) FIG. 2  
 = *Corynespora quercicola* Borowska, Acta Mycol. 11(1): 60 (1975)

SPECIMEN EXAMINED: on dead branches of unidentified plant, tropical forest of Bawangling, Hainan Province, China. 10 Dec 2009, J. Ma, HSAUP H5082 (duplicate HMAS 146083).

ANAMORPHIC FUNGI. Colonies effuse, blackish brown to black, hairy. Mycelium partly superficial, partly immersed in the substratum, composed of branched, septate, pale brown, smooth-walled hyphae, 2–4.5  $\mu\text{m}$  wide. Conidiophores differentiated, arising single or in groups, erect, straight or flexuous, unbranched, brown to dark brown, smooth, septate, 45–114  $\mu\text{m}$  long, 3–4  $\mu\text{m}$  wide, sometimes once or twice percurrent. Conidiogenous cells monotretic, integrated, terminal, cylindrical, brown, smooth. Conidia acrogenous, dry, in short acropetal chains, broadly ellipsoid to oblong, mainly 2-euseptate, rarely 1-euseptate, sometimes slightly constricted at the septum, smooth, polar cells pale brown, middle cell brown to dark brown, 13–21  $\mu\text{m}$  long, 6–7  $\mu\text{m}$  wide in the widest part.

NOTES: This is the first report of this species in China. The conidia of the specimen examined are somewhat longer (13–21  $\mu\text{m}$  vs. 12–18  $\mu\text{m}$ ) than those

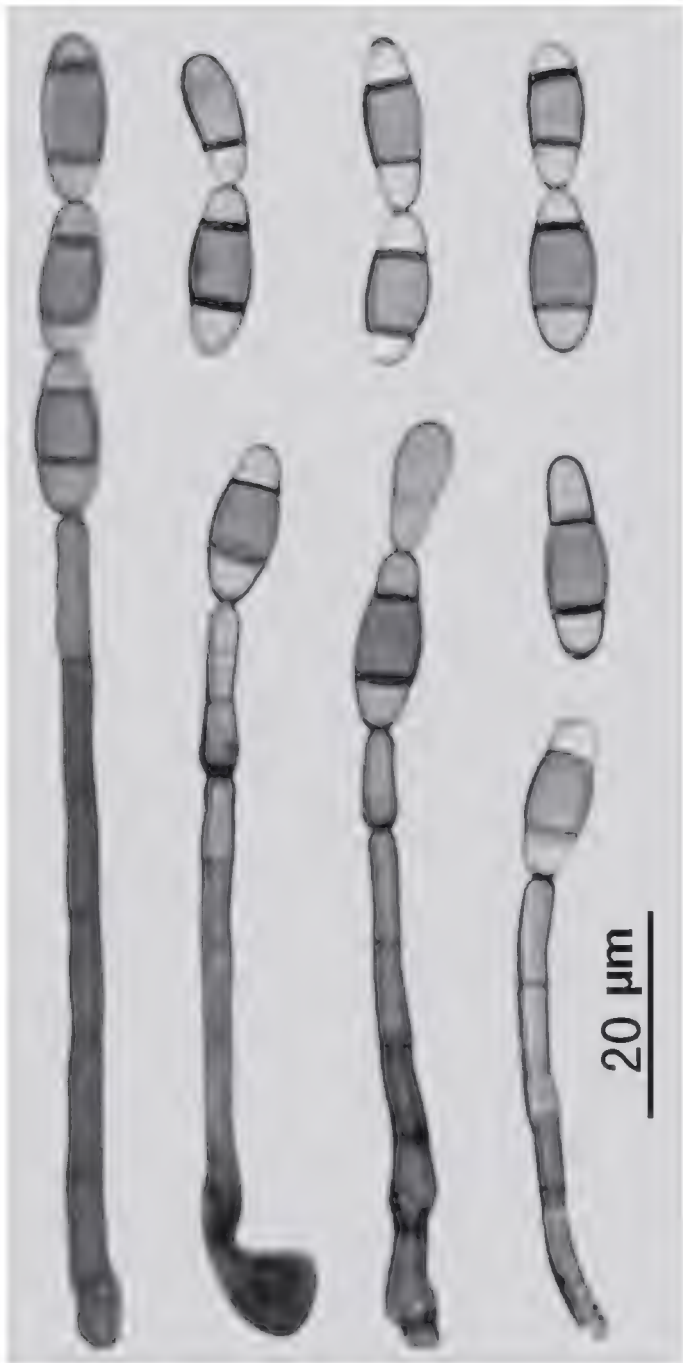


FIG. 2. *Corynesporopsis quercicola*. Conidiophores and conidia.

of the type specimen described by Kirk (1981a). This species has been recorded from Russia, Poland, United Kingdom, and Cuba. *Corynesporopsis quercicola* somewhat resembles *C. biseptata* (Morgan-Jones 1988) in conidial shape and septation but has smaller ( $13\text{--}21 \times 6\text{--}7 \mu\text{m}$  vs.  $18\text{--}33 \times 7\text{--}9 \mu\text{m}$ ) versicolored conidia.

*Corynesporopsis indica* P.M. Kirk, Mycotaxon 17: 405 (1983)

FIG. 3

SPECIMEN EXAMINED: on dead branches of unidentified plant, tropical forest of Baomeiling, Hainan Province, China. 9 Dec 2009, J. Ma, HSAUP H5274–1 (duplicate HMAS 146084).

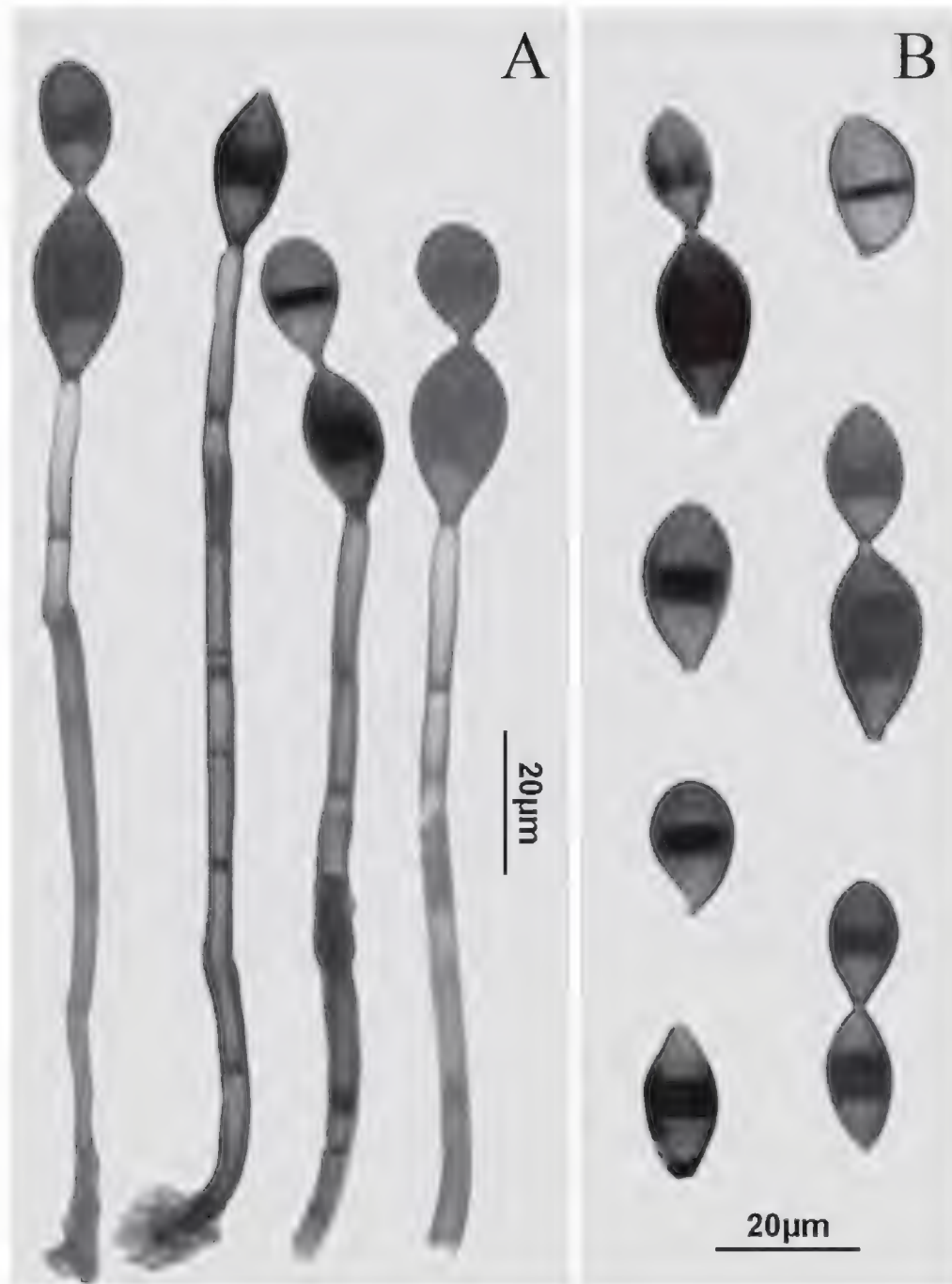


FIG. 3. *Corynesporopsis indica*. A. Conidiophores and conidia. B. Conidia.

ANAMORPHIC FUNGI. Colonies effuse, blackish brown to black, hairy. Mycelium mostly immersed in the substratum, composed of branched, septate, pale brown to brown, smooth-walled hyphae, 1.5–3 µm wide. Conidiophores differentiated, single or in groups, erect, straight or slightly flexuous, unbranched, brown to dark brown, smooth, septate, 67–172 µm long, 3–5.5 µm wide, sometimes swollen at the base. Conidiogenous cells integrated, terminal, monotretic, cylindrical, brown, smooth, sometimes with percurrent proliferation. Conidia acrogenous, dry, solitary or in acropetal chains of 2 or 3, ellipsoid to broadly obovoid, sometimes somewhat biconic, i with one indistinct median euseptum,



the septum usually obscured by a darkly pigmented band, smooth, dark brown to blackish brown, 16–27 µm long, 8–13.5 µm wide in the widest part.

NOTES: This species has not been previously recorded in China. The size range of conidia and conidiophores in our specimen overlaps well with that of the type specimen described by Kirk (1983), and other features of this taxon also match those of the original species. *Corynesporopsis indica* is unique within the genus in its ellipsoid to obovoid, 1-septate conidia with the septum usually obscured by a band of pigment.

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## MYCOTAXON

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**Black mildew fungi (*Meliolaceae*) associated with  
*Schinus terebinthifolius* (Brazilian pepper tree)  
in Brazil**

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**Abstract** — *Meliola chilensis*, *M. rhois* var. *africana*, and *Irenopsis schini-terebinthifolii* sp. nov. are described from the leaves of *Schinus terebinthifolius* (*Anacardiaceae*). Illustrations and a key to all *Meliolaceae* known to associate with species belonging to the genus *Schinus* are provided.

**Key words** — *Ascomycetes*, Atlantic forest, biodiversity, taxonomy, tropical fungi

## Introduction

*Schinus terebinthifolius* Raddi (*Anacardiaceae*), the Brazilian pepper tree (known in Brazil as “aroeira”), is a small sized plant widely distributed in Brazil, Argentina, and Paraguay. Introduced to many other tropical and sub-tropical regions as an ornamental or spice source, the Brazilian pepper has invaded natural ecosystems and provoked serious disruptions of such natural areas. Now regarded as one of the worst invasive plant species in Florida, Hawaii, and New Zealand (Ferriter 1997), for several decades it has been a target of biological control programs by using insects as its natural enemies (Cuda et al. 2006).

Surveys of and research on fungal pathogens of *S. terebinthifolius* have been only recently initiated in Brazil. A partial result of such surveys published by Faria et al. (2008) has revealed a significant diversity of pathogenic or purported pathogenic fungi associating with *S. terebinthifolius*. Some of these, such as *Septoria* sp., have clear potential for use in biological control of the

Brazilian pepper tree (Faria et al. 2008). The present publication deals with a group of species in the fungal family *Meliolaceae* collected on *S. terebinthifolius*. Although they clearly show no potential for biocontrol, they do represent mycological novelties.

The *Meliolaceae* includes approximately 1980 species, of which most are from the tropics (Kirk 2008). The main genera in this family are *Amazonia* Theiss., *Appendiculella* Höhn., *Asteridiella* McAlpine, *Irenopsis* F. Stevens, and *Meliola* Fr. (Hansford 1961). Meliolaceous fungi produce black colonies on the host surface and hence are known as black mildews. They have little economic importance even when attacking cultivated plants since the disease severity is generally low (Sabulal et al. 2006, Hosagoudar et al. 1997). In some cases, particularly when the host-species is an ornamental plant and black mildew colonies are abundant on it, this may harm the appearance of the plant as reported for *Asteridiella pittieri* (Toro) Hansf. attacking *Duranta repens* L. (Pereira et al. 2006).

Three distinct black mildew taxa were found during the survey of the mycobiota on *S. terebinthifolius*. Even not useful for biocontrol, their potential as scientific novelties justified further investigation. There is an obvious need to broaden the knowledge of the *Meliolaceae* in Brazil, as the group has been largely neglected by Brazilian mycologists and little has been published on this fungal group in Brazil in contrast to the large number of novel taxa in the *Meliolaceae* described from other tropical countries (Crane & Jones 2001, Hosagoudar & Shiburaj 2002, Song & Li 2004, Biju et al. 2005, Rodriguez & Piepenbring 2007).

## Materials and methods

The mycobiota on *S. terebinthifolius* was intensively surveyed during two different periods from September 2001 to May 2003, concentrated at first in a small part of southeastern Brazil in areas of the states of Rio de Janeiro, São Paulo, and Minas Gerais and later expanding to include also Espírito Santo, Paraná, Santa Catarina, and Rio Grande do Sul and additional ad hoc collections in the state of Pernambuco from 2008 to 2010. Foliage of *S. terebinthifolius* bearing black mildew colonies was collected, observed while still fresh, and then dried in a plant press. Samples were examined under an Olympus SZX7 stereomicroscope. Representative structures were either scraped with a scalpel or removed with an adhesive tape and mounted in lactophenol. Fungal structures were measured, photographed, and drawn using an Olympus BX 51 light microscope equipped with an Olympus e-volt 330 digital camera and a drawing tube. Representative specimens were deposited in the local herbarium at the Universidade Federal de Viçosa (Herbarium VIC).

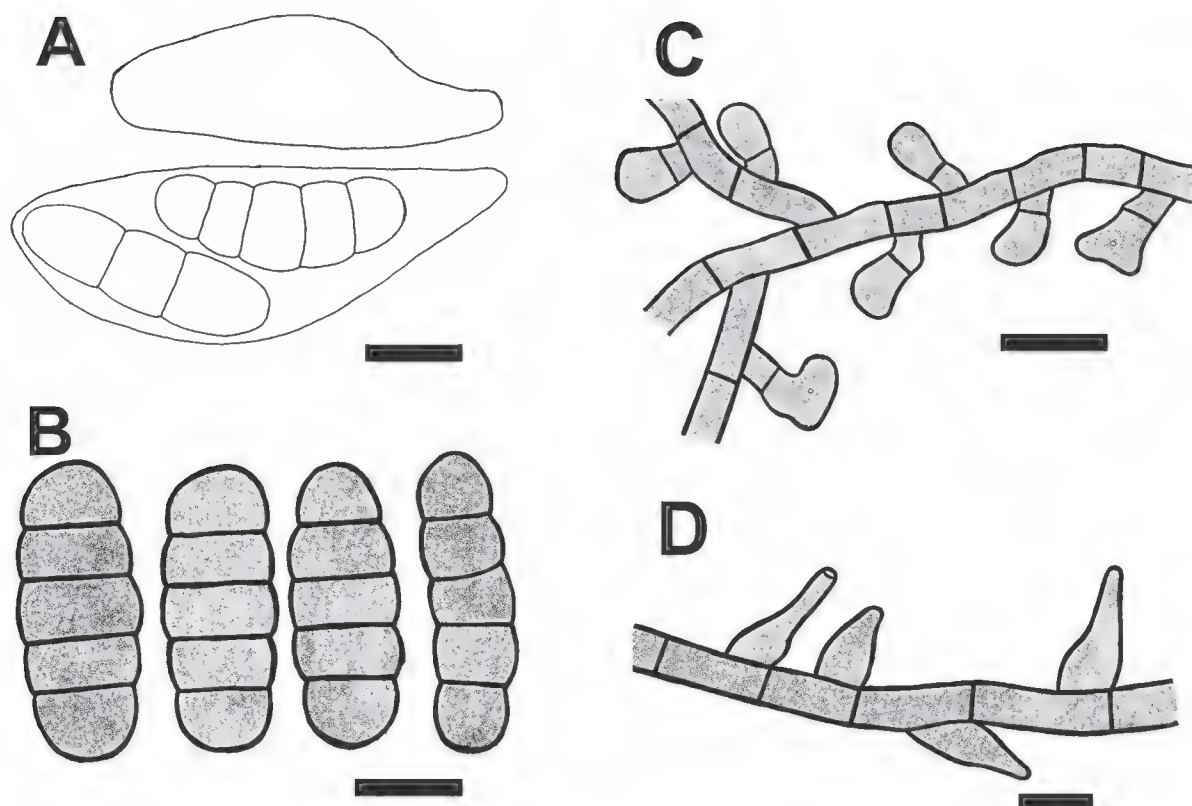


PLATE 1. *Meliola chilensis*. A. Asci and immature ascospores. B. Ascospores. (Bar = 20μm).  
C. Appressoria on hypha. D. Conidiogenous cells on hypha. (Bar = 20μm).

## Taxonomy

*Meliola chilensis* Speg., Bol. Acad. Nac. Ci. 25: 41 (1921)

PLATE 1

SPECIMENS EXAMINATED: on leaves of *Schinus terebinthifolius*, BRAZIL, Minas Gerais, Heliadora, 08 July 2008, D.M. Macedo (VIC 31323); São Brás do Suaçuí, 28 October 2009, D.M. Macedo (VIC 31336); Barbacena, 28 October 2009, D.M. Macedo (VIC 31337); Paraná, Bacaetava, 09 July 2008, D.M. Macedo (VIC 31322).

Colonies amphigenous, mostly epiphyllous, black, dense, subvelvety, 0.6–2.7 mm diam. Hyphae crooked, composed of dark brown septate hyphae, cells 15–26.5 × 7.5–9 μm, branching alternate to irregular at acute to irregular angles, producing appressoria and conidiogenous cells. Appressoria alternate, sub-antrorse, straight to bent; stalk cells cylindrical to cuneate, brown, 7.5–10 × 7.5–9 μm; head cell cylindric-clavate, straight to bent, entire, sometimes rounded-angulose to sublobate, brown or reddish brown, 14–20 × 13–19 μm. Conidiogenous cells (phialides) borne on a separate mycelia branch, opposite to alternate, ampulliform, brown, 11–15 × 5–7 μm. Mycelial setae numerous, scattered, straight to slightly flexuous, 7–12 septate, simple, apex obtuse, dark brown, 332–550 × 7.5–10 μm. Perithecia in a central group, black, globose, verrucose, 213–345 μm diam. Asci evanescent. Ascospores oblong, hyaline

when inside the ascus, becoming brown with age, rounded at the tips, 4-septate, constricted at the septa, dark brown,  $46\text{--}52 \times 15\text{--}20\ \mu\text{m}$ .

ADDITIONAL DESCRIPTION: Hansford (1961: 470).

BRAZILIAN DISTRIBUTION: Paraná and Minas Gerais (Brazil).

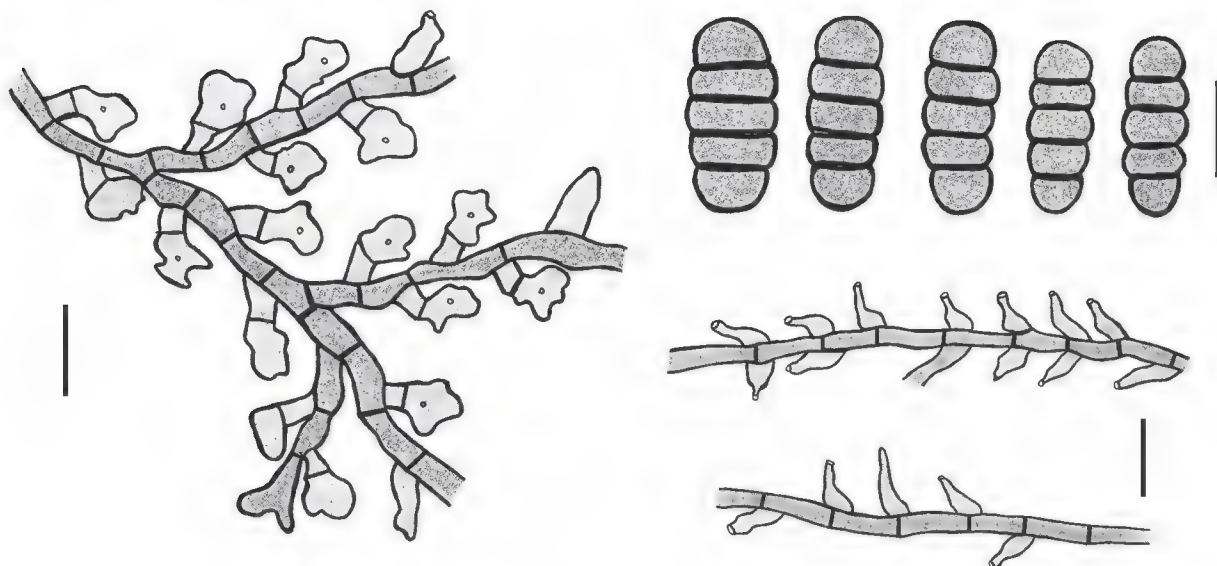


PLATE 2. *Meliola rhois* var. *africana*. A. Ascospores (Bar =  $20\mu\text{m}$ ). B. Appressoria on hypha (Bar =  $20\ \mu\text{m}$ ). C. Conidiogenous cells on hyphae (Bar =  $30\mu\text{m}$ ).

*Meliola rhois* var. *africana* Hansf., Sydowia 9: 75, 1955

PLATE 2

SPECIMEN EXAMINED: on living leaves of *Schinus terebinthifolius*, BRAZIL, Rio de Janeiro, Mury, 11 April 2008, D.M. Macedo (VIC 31320).

Colonies amphigenous, mostly epiphyllous, black, dense, velvety, cells 3–26 mm diam. Hyphae almost straight to sinuous, composed of dark brown septate hyphae, cells  $12.5\text{--}25 \times 7.5\text{--}9\ \mu\text{m}$ , branching usually alternate at acute angles, producing appressoria and conidiogenous cells. Appressoria alternate, more or less antrorse, straight or bent; stalk cells cylindrical to cuneate, brown,  $6\text{--}10 \times 6\text{--}7.5\ \mu\text{m}$ , head cell irregularly lobate, versiform, from elongate-clavate to broader than long, straight to variously bent, brown,  $12.5\text{--}22.5 \times 12.5\text{--}17.5\ \mu\text{m}$ . Conidiogenous cells (phialides) separate, opposite to alternate, ampulliform, brown,  $17.5\text{--}25 \times 7.5\text{--}9\ \mu\text{m}$ . Mycelial setae numerous, scattered, straight, 6–12 septate, simple, apex acute, dark brown,  $314\text{--}527 \times 8.5\text{--}10\ \mu\text{m}$ . Perithecia in a central group, black, globose, verrucose,  $243\text{--}354\ \mu\text{m}$  diam. Asci evanescent. Ascospores oblong to subellipsoid, hyaline when inside the ascus, becoming brown with age, rounded at the tips, 4 septate, constricted at the septa, dark brown,  $45\text{--}52.5 \times 14\text{--}19\ \mu\text{m}$ .

ADDITIONAL DESCRIPTION: Hansford (1961: 469).

BRAZILIAN DISTRIBUTION: Rio de Janeiro



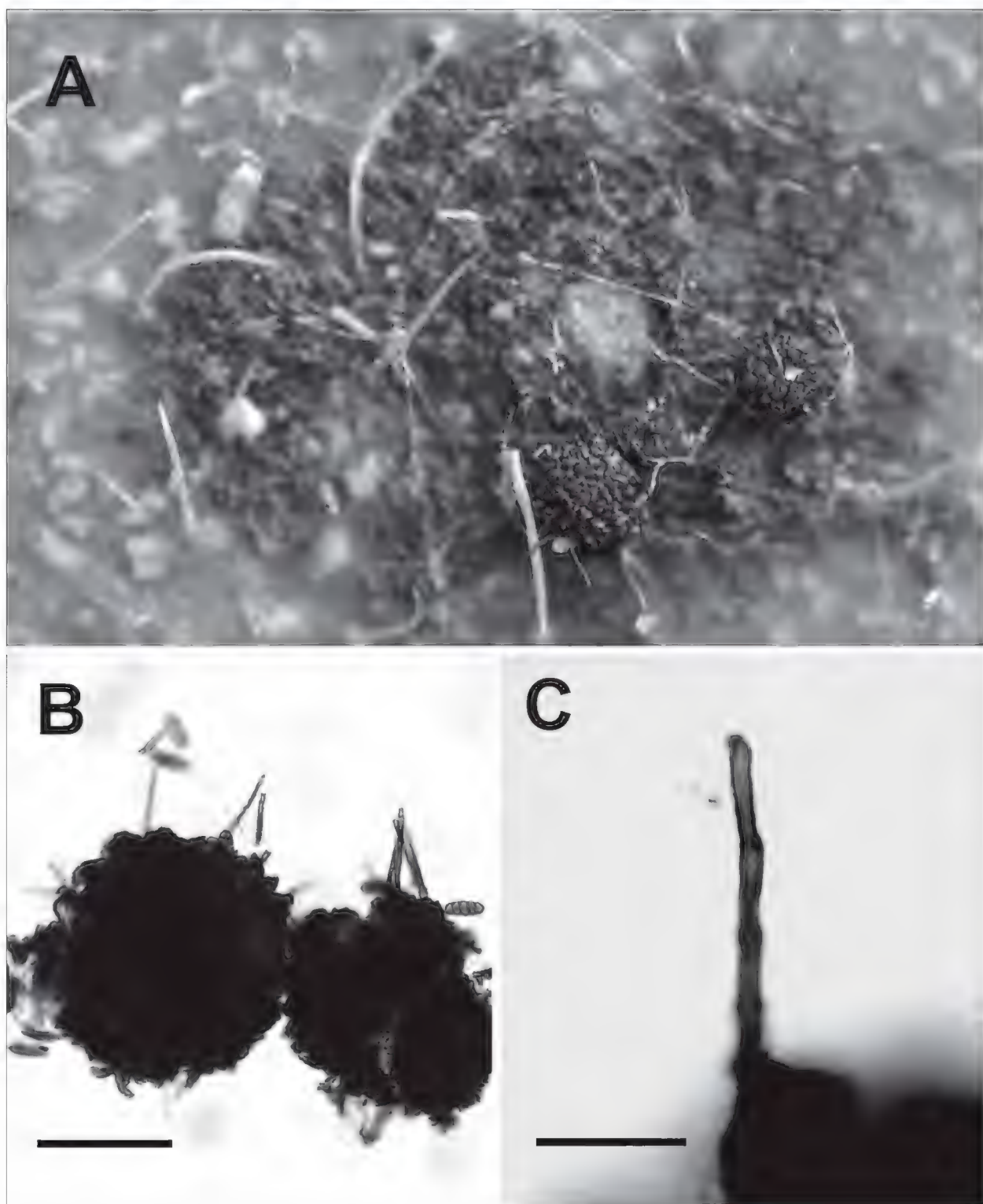


PLATE 3. *Irenopsis schini-terebinthifolii*. PLATE 3. A. Colony on leaf of *Schinus terebinthifolius*. B. Setose perithecia. C. Close-up of perithecial seta. (Bar = 25 µm).

***Irenopsis schini-terebinthifolii* D.M. Macedo & R.W. Barreto, sp. nov.**

MYCOBANK 18069

PLATES 3–4

*Differt a I. comocladiae* coloniae 0.4–2.1 cm; cellulae hyphales 15–40 × 7–9 µm. cellulae basalis appressoriis 6–11 × 6–10 µm; cellulae apicalis 12–19 × 11–16 µm. Cellulae conidiogenae oppositae, 19–21 × 6–8 µm. Perithecia 168–300 µm diam; setae peritheciales, 1–2 septatae, 64–140 × 5–7 µm; Ascosporae 40–50 × 15–23 µm.

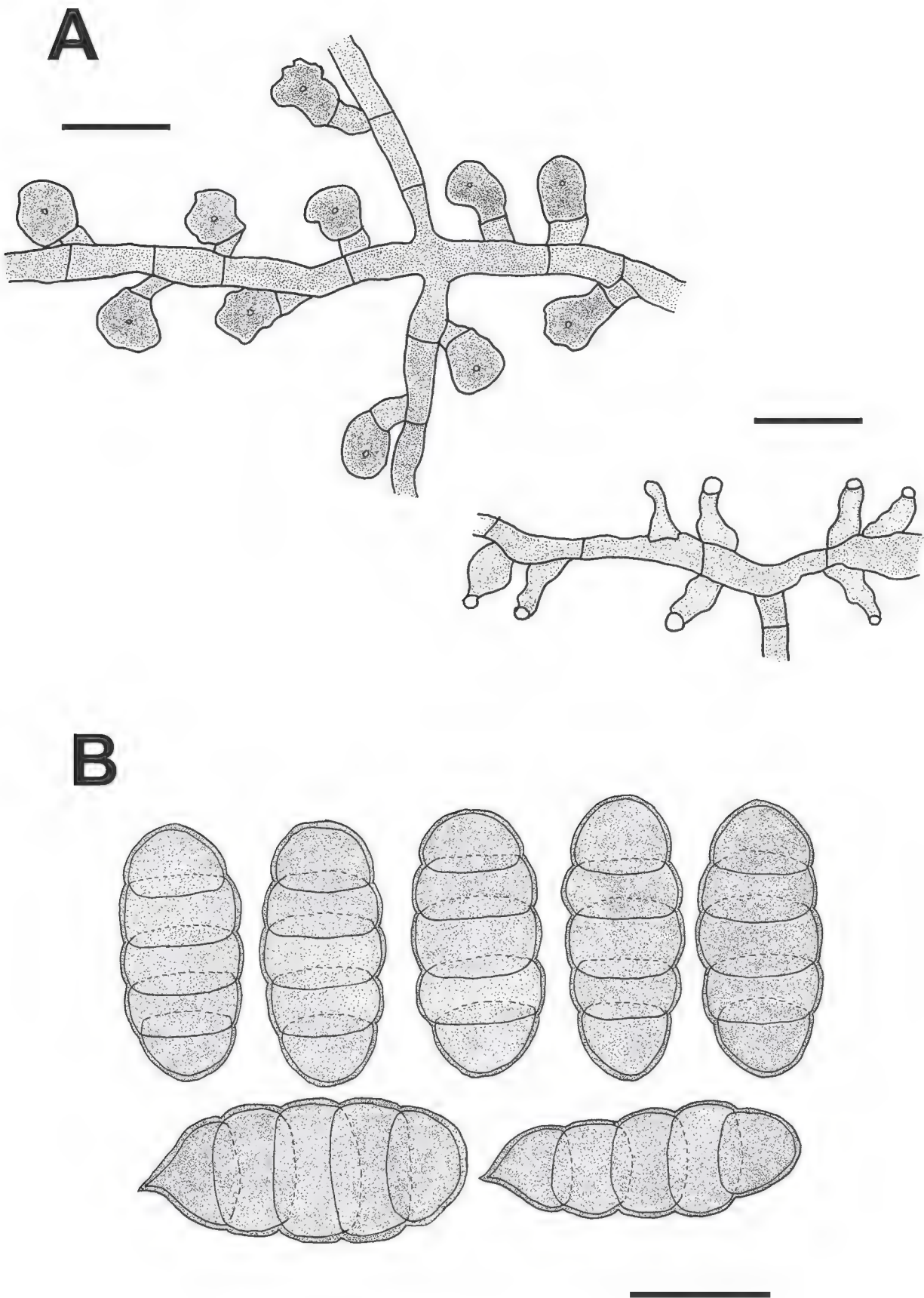


PLATE 4. *Irenopsis schini-terebinthifolii*. PLATE 4. A. Appressoria and conidiogenous cells on hyphae (Bar= 20  $\mu$ m). B. Ascospores (note two spores bearing pointed cells on left (Bar = 45  $\mu$ m)).

TYPE: on living leaves *Schinus terebinthifolius* (Anacardiaceae), D. M. Macedo, Casimiro de Abreu, Rio de Janeiro, Brasil (**holotype** - VIC 31318).

ETYMOLOGY: The epithet refers to the host plant, *Schinus terebinthifolius*

Colonies amphigenous, mostly hypophyllous, confluent, black, dense, scattered, cells 0.4–2.1 mm diam. Hyphae straight, almost straight to undulate, composed of dark brown septate hyphae, cells  $15\text{--}40 \times 7\text{--}9\text{ }\mu\text{m}$ , branching opposite at acute or wide angles, producing appressoria and conidiogenous cells. Appressoria alternate, antrorse, bent,  $18\text{--}30\text{ }\mu\text{m}$  long; stalk cells cylindrical to cuneate, brown,  $6\text{--}11 \times 6\text{--}10\text{ }\mu\text{m}$ , head cell irregular, entire to rounded-angulose, brown to reddish brown,  $12\text{--}19 \times 11\text{--}16\text{ }\mu\text{m}$ . Conidiogenous cells (phialides) mixed with appressoria, opposite, conoid to ampulliform, brown,  $19\text{--}21 \times 6\text{--}8\text{ }\mu\text{m}$ . Perithecial setae straight, 1–2 septate, simple, apex obtuse, dark brown,  $64\text{--}140 \times 5\text{--}7\text{ }\mu\text{m}$ . Perithecia black, scattered, globose,  $168\text{--}300\text{ }\mu\text{m}$  diam. Asci evanescent. Ascospores oblong to subellipsoid, end cells often pointed at apex, hyaline when inside the ascus, becoming brown with age, rounded at the tips, 4-septate, constricted at the septa, dark brown,  $40\text{--}50 \times 15\text{--}23\text{ }\mu\text{m}$ .

DISTRIBUTION: Rio de Janeiro and Minas Gerais (Brazil).

ADDITIONAL SPECIMENS EXAMINED: on living leaves of *Schinus terebinthifolius*, **BRAZIL**, Rio de Janeiro, parque Nacional de Jurubatiba, 9 April 2008, D.M. Macedo (VIC 31319); Casimiro de Abreu, 11 April 2009, D.M. Macedo (VIC 31321); Minas Gerais, Ponte Nova, 22 August 2008, D.M. Macedo (VIC 31325); Padre Viegas, 22 August 2008, D.M. Macedo (VIC 31326); Catas Altas, 24 August 2008, D.M. Macedo (VIC 31226); Antonio Pereira, 23 August 2008, D.M. Macedo (VIC 31340); Lambari, 23 March 2009, D.M. Macedo (VIC 31334).

COMMENTS – The three meliolaceous fungi collected on *S. terebinthifolius* clearly belong to *Irenopsis* and *Meliola*. The latter is easily separated from *Appendiculella*, *Asteridiella*, and *Irenopsis* by mycelial setae; *Asteridiella* has no setae, *Appendiculella* has perithecia bearing larviform appendages, and *Irenopsis* has setose perithecia (Hansford, 1961).

Forty-two species and 7 infraspecific taxa of *Meliolaceae* are known on members of *Anacardiaceae*. Of these, 38 species and 7 infraspecific taxa belong to *Meliola* (Hansford 1961, Hosagoudar 1996, Hosagoudar & Archana 2009). The following *Meliolaceae* taxa have been reported in association with members of *Schinus*: *Meliola chilensis*, *M. lanigera* Speg., *M. rhoina* Doidge, *M. rhoina* var. *schini* Hansf., *M. rhois* var. *africana* and *M. rhois* var. *lithraeae* Hansf. (Hansford 1961, Mafia et al. 2004, Farr & Rossman 2010, Mendes & Urban 2010). With the exception of *M. chilensis*, all have been reported from Brazil, but only *M. lanigera* was reported in association with *S. terebinthifolius*.

The first fungus described above fits well within the description of *M. chilensis*, a fungus originally known on *Schinus latifolius* (Gillies ex Lindl.) Engl. and *Schinus latifolius* var. *tomentosus* Fenzl from Chile. The second fungus clearly belongs to *M. rhois* var. *africana*, which has been reported on *Rhus glaucescens*

A. Rich. in Uganda and Congo, on *Protorhus longifolia* (Bernh.) Engl. in South Africa, on *Schinus dependens* Ortega in Brazil, and on *Schinus molle* L. in Argentina, Brazil, and Paraguay. Therefore, the two *Meliola* taxa described above represent first reports on *S. terebinthifolius*. There are often more than one species of black mildew associated with plants in the *Anacardiaceae*. For instance, *S. latifolius* is a host for both *M. chilensis* and *M. rhoina* var. *schini*, while *S. dependens* serves as host for *M. lanigera*, *M. rhoina*, and *M. rhois* var. *africana* (Hansford, 1961).

Only two *Irenopsis* species have been described on members of the *Anacardiaceae*: *I. comocladiae* (F. Stevens) F. Stevens and *I. portoricensis* F. Stevens (Hansford 1961, Farr & Rossman 2010, Mendes & Urben 2010). The new specimen referred to *S. terebinthifolius* is the first time an *Irenopsis* species has been reported on a member of the genus *Schinus*. *Irenopsis schini-terebinthifolii* is distinguished from *I. comocladiae* and *I. portoricensis* by its simple and straight perithecial setae, longer cells at the appressoria bases and larger ascospores.

**Key to *Meliolaceae* taxa associated with *Schinus* spp.**

- 1. Mycelium not setose ..... *Irenopsis schini-terebinthifolii*
- 1' Mycelium setose ..... 2
- 2. Perithecia dispersed over colony ..... *Meliola rhois* var. *lithraeae*
- 2' Perithecia in a central group on colony ..... 3
- 3. Setae grouped around perithecia ..... *M. rhoina* var. *schini*
- 3' Setae scattered over colony ..... 4
- 4. Setae broadly arcuate to flexuous ..... *M. lanigera*
- 4' Setae straight ..... 5
- 5. Appressoria cylindrical-clavate. .... *M. chilensis*
- 5' Appressoria otherwise ..... 6
- 6. Ascospores 33–45 × 14–18 µm ..... *M. rhoina*
- 6' Ascospores 45–50 × 20–22 µm ..... *M. rhois* var. *africana*

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## Morphology: still essential in a molecular world

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**Abstract** — Morphological characters have long served as the basis for mycological taxonomy. But with the advent of DNA sequence data, is morphology still useful? Will barcoding replace visual identification? Taxa in the *Dothideomycetes* serve to illustrate how molecular analyses have revised species relationships and higher-level systematics. *Aspergillus* species are now defined using a polyphasic approach with morphology assuming a lesser role. Sequence analyses likewise reveal that *Colletotrichum* species complexes once considered good morphological species now comprise many phylogenetically distinct species. Although *Phyllosticta* species concepts are less advanced, sequence data are expected to reveal new species in that genus as well. Molecularly supported higher taxa in *Dothideomycetes* often differ from those circumscribed by morphological characters. However, DNA barcodes, recently applauded as a magic formula for species identification, are yet to be determined for many genera, and too many GenBank sequences are wrongly named or contain sequencing errors. Thus, despite recent molecular advances, there is an unprecedented need for mycologists to return to the field, recollect species, and re-typify taxa with living cultures. Only after we obtain sequences from species and genera linked to properly named taxa will barcoding become successful.

**Key words** — anamorph, molecular phylogenetics, teleomorph, traditional taxonomy, typification

## Introduction

Morphology has been the basis of nearly all fungal taxonomic studies. Numerous books and monographs use morphology alone to separate families, genera,

and species. Classical texts such as *MARINE MYCOLOGY, THE HIGHER FUNGI* (Kohlmeyer & Kohlmeyer 1969), *GENERA OF HYPHOMYCETES* (Carmichael et al. 1980), and *THE COELOMYCETES* (Sutton 1980) are archetypal examples. Numerous important higher-level taxonomic texts have also been published using morphology for all class, ordinal, and familial placements. Texts such as *A RE-EVALUATION OF THE BITUNICATE ASCOMYCETES WITH KEYS TO FAMILIES AND GENERA* (von Arx & Müller 1975) and *PRODROMUS TO CLASS LOCULOASCOMYCETES* (Barr 1987) are classic examples.

Clearly morphology has underpinned taxonomic studies. In many other areas of fungal biology, it is essential to establish correct names and until recently there has been no way to identify a fungus without using morphological characters. Thus most fungal biochemistry, biotechnology, bioremediation, physiology, and plant pathology studies have cited species named after the fungi were identified through morphology (e.g. novel compounds — Evidente et al. 2008; chitinase production — Souza et al. 2003; bioremediation — Launen et al. 1995; physiology of *Colletotrichum graminicola* — Ali 1962; checklist of disease associated microorganisms in northern Australia — Hyde & Alcorn 1993). Similarly, most ecological studies relied on morphology to identify fungal communities (e.g. soil fungi communities — Ali-Shtayeh & Jamous 2000; fungal succession — Duong et al. 2008; endophytes — Hyde & Soyong 2008).

The situation however, is rapidly changing. Monographs of many genera now almost entirely rely on molecular data, and increasingly more often morphology is being replaced by molecular study (e.g. Tejesvi et al. 2007, Aveskamp et al. 2010). Ecological studies may now completely ignore morphology and fungal communities are identified through analysis of environmental DNA (Seena et al. 2008, Curlevski et al. 2010). The identities of fungi used in population genetics, biotechnology, and even biochemical studies are now often checked using sequence data only.

The results of these changes are rarely questioned, let alone discussed, yet most mycologists would agree that these changes should be advantageous. In this paper we explore *Aspergillus*, *Colletotrichum*, and *Phyllosticta*, genera where sequence data have to some extent profoundly affected species understanding. Below we discuss the effect of sequence data on understanding higher taxonomic levels in the *Dothideomycetes* and illustrate some unsolved problems in the new system. The aim is neither to criticize the studies nor to degrade the outcome, but to point out the resulting changes and confusion so that the mycological community can deliberate how best to manage such changes to everyone's benefit.

## Phylogenetic methodology

Sequences were downloaded from GenBank and aligned using Clustal X. The alignment was optimized manually to allow maximum alignment and maximum sequence similarity. Gaps were treated as missing data. Phylogenetic analysis was carried out based on the aligned dataset by PAUP\* 4.0b10 (Swofford 2002). Ambiguously aligned regions were excluded from all analyses. Trees were inferred using the heuristic search option with TBR branch swapping and 1000 random sequence additions. Maxtrees were unlimited, branches of zero length were collapsed, and all multiple parsimonious trees were saved. Trees were figured in TreeView (Page 1996).

## Discussion

### *Aspergillus*, *Colletotrichum* and *Phyllosticta* – the process towards understanding a species

In many genera understanding what delimits a species has typically evolved from 1) a basic and relatively stable morphological concept (possibly including other characters such as cultural, growth rates, or mating), which often comprised species complexes, to 2) molecular revision where the morphological system starts to disintegrate and needs rethinking, and 3) a stabilized system based on molecular data with morphology taking a lesser role. Although eventually taxa may be identified solely using molecular data, in most genera this is decades away.

*Aspergillus* is advanced with respect to species delineation, mainly because it produces post harvest mycotoxins and valuable industrial chemicals (Geiser et al. 2007, Samson & Varga 2007). There has been a substantial increase in numbers of accepted taxa, with Rapier & Fennell (1965) recognizing 132 species, Geiser et al. (2008) estimating ~250 species, and Kirk et al. (2010) 266 species. Species delineation is based on a polyphasic approach with molecular data taking primary importance (Geiser et al. 2007). Multiple independent loci are now recommended when describing new species, particularly loci for which large datasets already exist, such as ITS,  $\beta$ -tubulin, calmodulin, actin, and RNA polymerase (Samson et al. 2007). All types are available in culture collections (Pitt & Samson 2000). Many species have now been sequenced for multiple genes and the understanding of species concepts in *Aspergillus* is advanced. Whole genomes have also been sequenced for at least eleven strains of nine species, with several others in the pipeline (Geiser et al. 2007; Samson, pers. comm.).

Sutton (1980) provided a practical key to 40 *Colletotrichum* species that provided a basic species identification text. Although often difficult to decide whether to key a fungus to one or another species, the key was convenient and

descriptions brief. Even after 27 years and >4000 *Colletotrichum* publications, Sutton's text served as a necessary and convenient tool for placing names on taxa. The first molecular data on *Colletotrichum* were published after 1990 (e.g. Bailey et al. 1996, Correll et al. 1993, Fabre et al. 1995); although the results were revealing, the data began to complicate species identification (Hyde et al. 2009a, b). There was, however, no attempt to stabilize species concepts in a formal way, so that sequences deposited in GenBank were unknowingly often wrongly named. Not until 2007–2008 were several *Colletotrichum* species epitypified (Shenoy et al. 2007, Cannon et al. 2008), thereby enabling comparisons of reference sequence data against data from fresh collections. This commenced the period of reconciling *Colletotrichum* species, especially in the difficult complexes. Recent studies have introduced 15 new species (most in the “*gloeosporioides*” species complex), epitypification of 14 *Colletotrichum* species, and generation of sequence data for ex-type cultures of 46 species (Hyde et al. 2009b; Damm et al. 2009; Prihastuti et al. 2009, 2010; Shivas & Yu 2009; Phoulivong et al. 2010; Yang et al. 2009, 2010; Wikee et al. 2011).

FIGURE 1 provides an example of the confusion that molecular data can produce. We generated the phylogram by downloading 41 GenBank ITS sequences, of which 25 were labeled *Colletotrichum gloeosporioides*. In FIG. 1 *C. gloeosporioides* epitype sequences cluster at the top of the tree, while clades containing putative *C. gloeosporioides* strains — some representing very distantly related species — are scattered throughout, illustrating the diversity of one species name in GenBank. Cai et al. (2009a) have estimated that >86% of the *C. gloeosporioides* names in GenBank considerably diverge from the epitype and are likely to represent other *Colletotrichum* species. As *C. gloeosporioides* represents a species complex comprising numerous diverse species, great care must be used when downloading sequences labeled as ‘*gloeosporioides*’ from GenBank. Ultimately, only sequence data from the epitype strain should be used to characterize the species.

Compared with *Aspergillus* and *Colletotrichum*, understanding *Guignardia* and its *Phyllosticta* anamorphs is less advanced. *Guignardia* comprises 335 records (Index Fungorum) and has no monograph, although species from various hosts have been reviewed (e.g. palms — Hyde 1995; *Podocarpus* — Crous et al. 1996). Van der Aa & Vanev (2002) accepted 141 species based on cultural and morphological characteristics in their monograph on *Phyllosticta*. As very few living types appear to exist in these genera, Wulandari et al. (2009) compared their new species causing tan spot of pomelo in Asia with many questionably labeled *Phyllosticta* sequences from GenBank. D.M. Lam & N. Wulandari (unpublished) also sequenced many *Guignardia* and *Phyllosticta* strains from CBS, but as few represented type strains, their conclusions were limited and may never be published. There is a need to designate epitypes



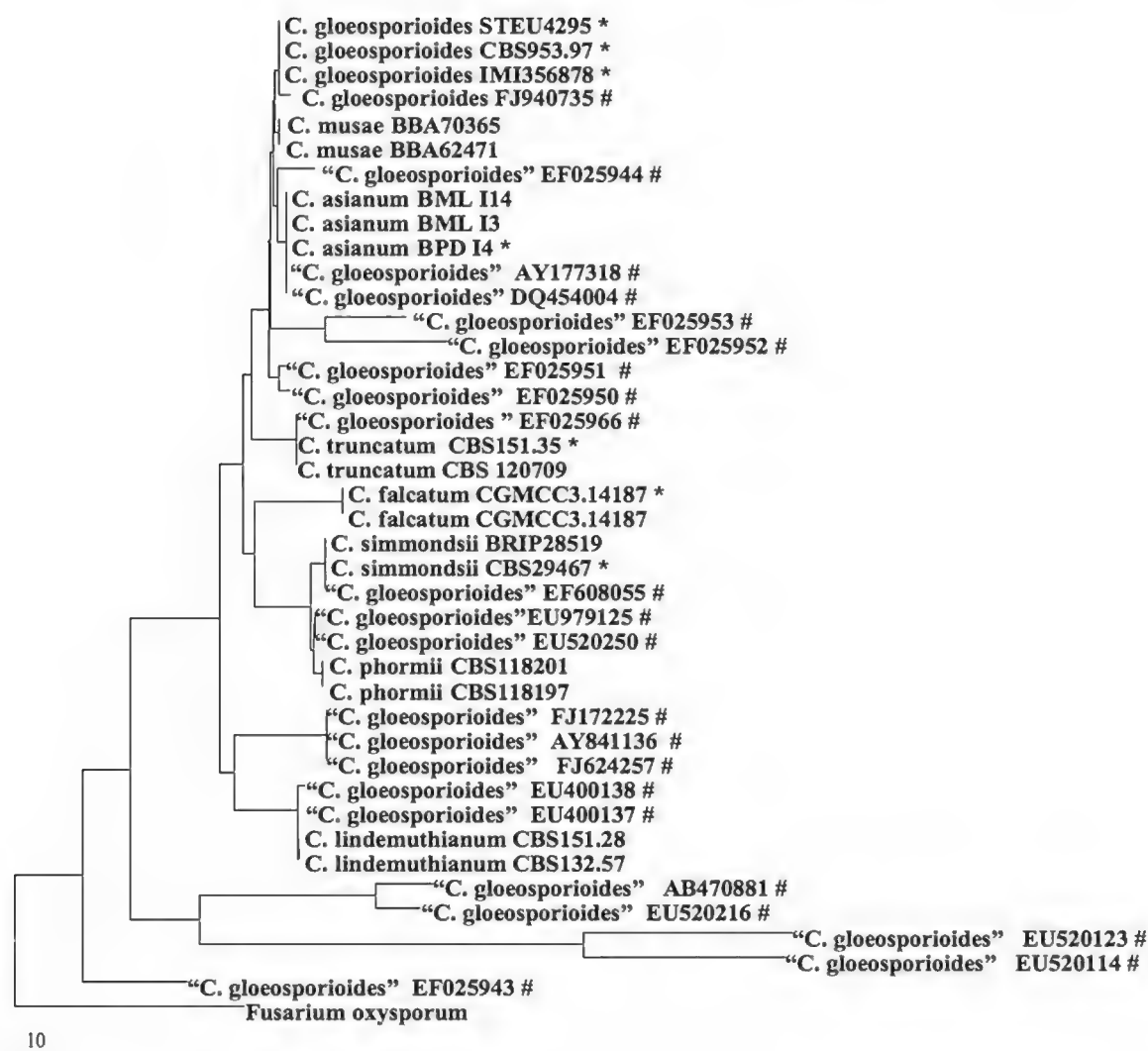


FIG. 1. Maximum parsimony phylogram generated from ITS sequence analysis of "*Colletotrichum gloeosporioides*" downloaded from GenBank with other related taxa. Data were analysed with random addition sequence, unweighted parsimony, and treating gaps as missing data. # indicates ITS sequences of "*Colletotrichum gloeosporioides*" downloaded from GenBank; \* indicates sequences derived from ex-type cultures.

for species of *Phyllosticta* and the teleomorph *Guignardia*, so that a clear understanding of the status of species and their biological relationships can be obtained.

*Guignardia mangiferae* A.J. Roy offers a second example of confusion resulting from molecular data (FIG 2). This name has been extensively applied to an endophyte isolated by Rodrigues et al. (2004); many putative *G. mangiferae* strains were used by Wulandari et al. (2009). However, no type of *G. mangiferae* can be found (Wulandari, pers comm.) nor has it ever been epitypified. Thus this recent name has been used arbitrarily for endophytic strains producing obtrullate ascospores. The obtrullate ascospore type, however, can be found in numerous species (e.g. *G. eucalyptorum* Crous, *G. smilacis* A.J. Roy, *G. graminea* Lobik) and most likely comprises a species complex that could

have a much older name. In FIG. 2 we downloaded a selection of *G. mangiferae* labeled strains from GenBank to illustrate the diversity the name represents. It is therefore unwise to name a *Guignardia* or *Phyllosticta* species based solely on sequence similarity with a GenBank sequence.

The above examples serve to illustrate how molecular data can resolve species understanding in some plant pathogenic genera yet pose challenges in interpretation. We should remember that many previous studies likely applied incorrect names to their organisms. Type cultures must be sequenced, and where no such cultures exist, fresh collections are needed. Both type cultures and fresh collections should be fully characterized using morphology, sequence analyses, and other polyphasic approaches. Only by using such methods can we begin to understand genera and their individual species complexes. Such understanding now exists for *Aspergillus* and *Penicillium*, is advanced in *Fusarium*, is progressing in *Colletotrichum*, and has only begun in *Guignardia*/*Phyllosticta* and *Pestalotiopsis*. The simple message is that although molecular data may eventually identify taxa in these genera, an enormous concerted effort is needed to recollect, morphologically characterize, epitypify, sequence, analyze, and combine all data with other polyphasic characters before we will make any real progress in understanding species in these important genera. It is also suggested that NCBI should rename an entry if there are sufficient evidences supporting to do so.

### **The *Loculoascomycetes***

AFTOL (All Fungi Tree of Life) aimed to find natural classifications for fungi based on multi-locus phylogeny, rather than visual, relationships (Schoch et al. 2006). The project made considerable progress towards understanding fungi at the higher levels, particularly in the basidiomycetes. Classes of fungi are similarly better resolved in the ascomycetes, although the *Dothideomycetes* offer a good example where molecular analyses have resulted in uncertainty, especially at the family level.

The issue of STUDIES IN MYCOLOGY (Schoch et al. 2009) devoted to the *Dothideomycetes* resolved many problems at the higher taxonomic levels (order, family) but may have created more confusion than intended. What classical mycologists such as J.A. von Arx, E. Müller, and M.E. Barr previously considered to be orders and families and the characters they used to diagnose such (von Arx & Müller 1975; Barr 1987) are, in many cases, no longer usable. Unfortunately, although molecular data can place taxa at the family and in some cases generic levels, there has been little effort made in attempting to correlate phylogeny with phenotypes (Suetrong et al. 2009, Zhang et al. 2009a).

For example, the *Lophiostomataceae* and *Trematosphaeriaceae* cluster as separate families and contain elements that can be linked by very few characters

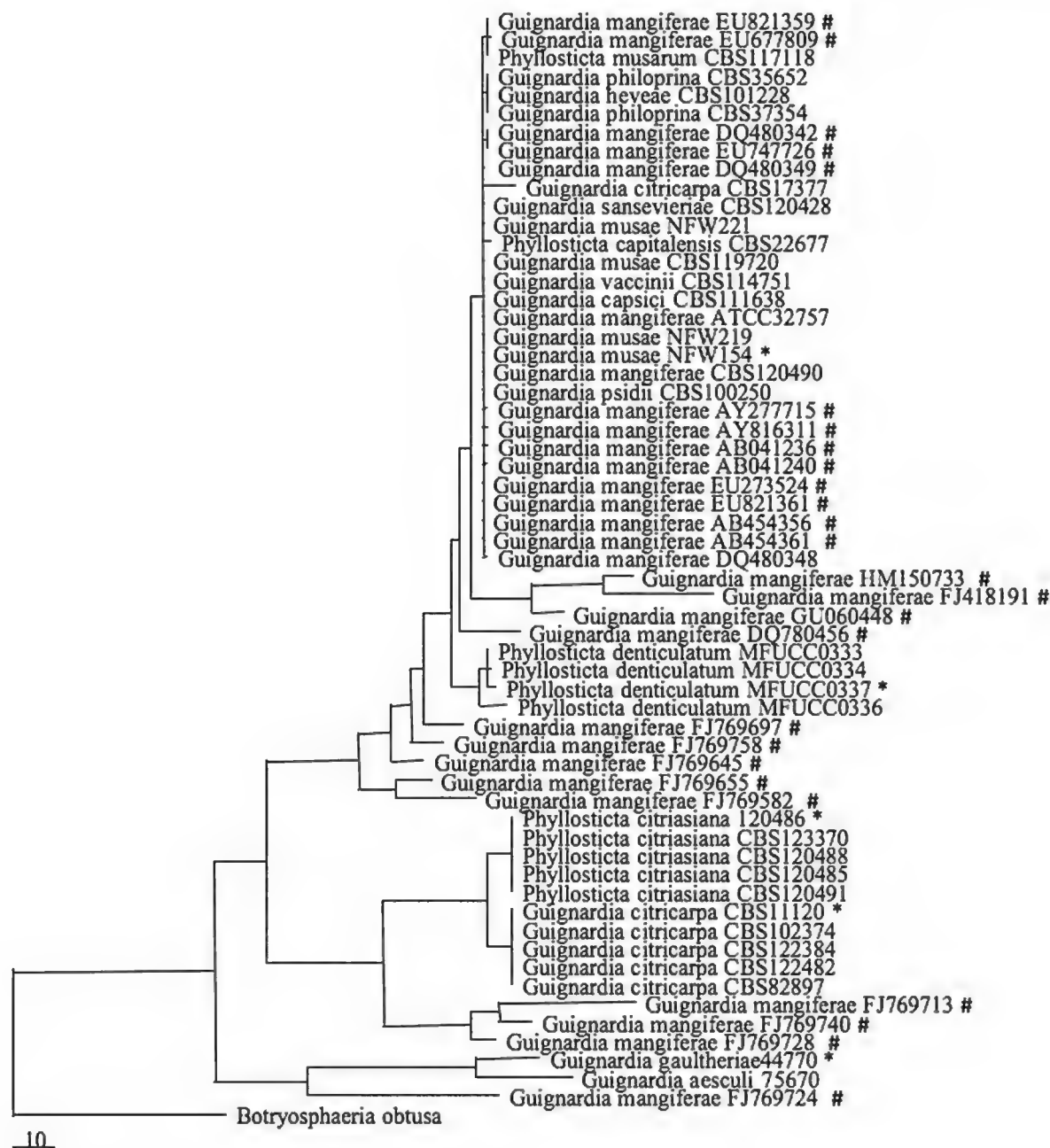


FIG. 2. Maximum parsimony phylogram generated from ITS sequence analysis of “*Guignardia mangiferae*” downloaded from GenBank with other related taxa. Data were analysed with random addition sequence, unweighted parsimony, and treating gaps as missing data. # indicates ITS sequences of “*Guignardia mangiferae*” downloaded from GenBank; \* indicates sequences derived from ex-type cultures.

— the same characters found in other families. The *Lophiostomataceae* include *Lophiostoma*, some species placed in *Thyridaria*, and a new genus *Misturatosphaeria* (Mugambi & Huhndorf 2009; Zhang et al. 2009a,b). *Lophiostoma* species are characterized by ascomata that are erumpent with slot- or slit-like ostioles and may have raised flanges (Holm & Holm 1988), while in *Misturatosphaeria* ascomata are erumpent to superficial with often raised rounded apices and ascospores are phragmosporous or dictyosporous (Mugambi & Huhndorf 2009). Dictyosporous ascospore types are found throughout the

*Dothideomycetes* but not — until now — within *Lophiostomataceae*. At the moment, there is a distinct lack of defining characters that can be used for this family. Mugambi & Huhndorf (2009) themselves state, “despite morphological differences of *Misturatosphaeria* from other lophiostomataceous fungi, we feel justified in placing it in *Lophiostomataceae* at this point due to the strong support received in their analysis.”

*Tetraplosphaeriaceae* (Tanaka et al. 2009) is basal to most families in *Pleosporales* and yet previous classification systems would have probably placed the species in *Astrosphaeriella* (Hyde et al. 2000). The main distinguishing characters of the family are the *Tetraploa*-like anamorphs; however the ascomata (immersed or superficial), pseudoparaphyses (cellular or trabecular), and ascospore (fusiform to cylindrical, 1–3-septate, hyaline or pale brown) forms are found throughout the *Dothideomycetes*. Therefore if a researcher encounters the teleomorph stage only, it would be difficult to use morphology to place the taxon, even at the family level, unless the characters are identical to an existing species in the literature.

In other groups in the *Dothideomycetes* there are so few sequences available that phylograms reveal very little information concerning the species at any level. This is true of taxa in the *Capnodiaceae* and *Microthyriaceae* and in numerous genera (e.g. *Muyocopron*, *Trichodelitschia*) (see Boehm et al. 2009, Schoch et al. 2009).

What is the way forward? Many sequences used in the issue of STUDIES IN MYCOLOGY on the *Dothideomycetes* are linked to cultures from poorly documented taxa while only a few are linked to type material. This will create doubt in the minds of readers because generic types must be used in such analyses. Again, a concerted effort is needed to recollect, document characters, isolate, and deposit herbarium materials and/or living cultures. In this way we will have accurately documented morphological characters that are linked to sequence data of accurately named species; only then can we confidently start to understand relationships in *Dothideomycetes* and be confident in the conclusions arising from combined morphological and molecular classifications.

### Linking anamorphs to teleomorphs

There has been much expectation amongst mycologists that molecular analyses of anamorphic fungi will be able to link them to teleomorphs or at least provide an idea of their positions in the *Ascomycota* (Shenoy et al. 2006, 2007). Several studies have shown that morphological characters traditionally used to delimit anamorphic fungi are less informative in inferring fungal phylogenies. For example, in traditional taxonomy morphologically well-defined genera such as *Chalara* and *Sporidesmium* appear to be highly polyphasic (Shenoy et al. 2006, Cai et al. 2009b). Re-evaluation of the evolutionary significance of anamorphic



characters should therefore be carried out to 'rebuild' morphological classification. Morphology will then once again become important for identifying species, provided type specimens and derived cultures have been used in the reconstruction. If unavailable, the fungus should be interpreted by a freshly collected material from original hosts and localities, accurate documentation, isolation, sequencing, and deposition in herbaria as epitypes with living ex-type cultures. Only in this way will an accurate understanding of the natural placement of anamorphs in the teleomorphic scheme be achieved.

### **Barcoding and GenBank difficulties and solutions**

There are important initiatives to barcode the fungi (Santamaria et al. 2009, Seifert 2009). However, we feel that the benefit gained from large scale sequencing of fungal isolates will be diluted if sequence data from too few properly named taxa or types are deposited in public databases. As illustrated by Figs 1–2, the lack of sequences with reliably applied names in public databases would make barcoding currently unworkable. This deficiency must be corrected at the same time as barcoding takes place. As the type specimens and derived type cultures are not always available, there needs to be a concerted effort by mycologists to go back to the field and recollect the fungi. Taxonomic experts must carefully name those fungi and where possible designate epitypes with derived living cultures. Once we obtain sequences from species and genera that are linked to properly characterized taxa, we can really start to understand the fungi. Only then will barcoding work. These approaches will be useful in a few fungal studies as data obtained from molecular analysis of environmental samples, linking of anamorphs and teleomorphs, and the proper naming of species in biochemistry, pathology, and biotechnology research publications become precise.

### **Concluding remarks**

Fungal systematics has irreversibly stepped into the phylogenetics era. Molecular diagnosis through barcoding is favored by most researchers because it seemingly provides an easy and quick assessment of the fungus at hand and does not require years of training. This, however, does not exclude morphology from modern systematics, as morphological characters are the most easily accessible. The characters used to define species, genera, families, and orders nonetheless need reevaluation in light of sequence generated phylogenetic relationships. Morphological characters would then be used in agreement with new classification schemes and thus correspond to the natural phylogeny. The success of molecular diagnosis and barcoding, however, largely depends on comparing sequence data from type specimens. Most fungal names lack living type specimens and cannot be sequenced. There is consequently an



urgent need to epitypify all such fungi and deposit living ex-type cultures and derived sequence data in public culture collections and databases. Mycologists must go back to field and recollect important species and generic types and re-characterize these taxa using a polyphasic approach. Incorporating morphology is essential for establishing species concepts and higher taxonomic frameworks. Until much more data has been generated from types and many more accurately named species are deposited in public databases, confusion will remain. To eliminate the confusion, morphology is not only not outdated but is a necessity.

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## MYCOTAXON

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***Masseella flueggeae* on *Flueggea virosa*,  
a new record for Pakistan**

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**Abstract** — *Masseella flueggeae* on *Flueggea virosa* is reported as a new record for Pakistan. This is the first report of the genus *Masseella* from this country, raising the number of rust genera known from Pakistan to twenty-two.

**Key words** — *Euphorbiaceae*, macrocyclic rust, Neelum valley

## Introduction

*Flueggea virosa* is a dioecious, multi-stemmed, fast-growing bushy shrub in the *Euphorbiaceae*. It is common in deciduous woodlands and on forest margins, along rivers, and in rocky areas and is widely distributed in Asia, Africa, and Australia. In Pakistan, it is found in Sindh, the Kaghan Valley, and Kashmir (Stewart 1972). In the Neelum Valley, Azad Kashmir, this plant was found heavily infected by a rust fungus that belongs to an interesting rust genus, *Masseella* Dietel.

*Masseella* was erected by Dietel (1895) based on *M. capparis* (Cooke) Dietel [as “*capparidis*”] to accommodate a rust on *Flueggea virosa* in India and named after the famous English mycologist G.E. Masee (Cummins & Hiratsuka 2003). This genus is subtropical in distribution and restricted to the warm regions of Asia in the Philippines as well as South Africa. All species of *Masseella* parasitize members of the family *Euphorbiaceae* and are macrocyclic and autoecious (Thirumalachar 1943, Singh & Singh 1967). *Masseella flueggeae* on *Flueggea virosa* was described from the Philippines by Sydow & Petrak (1928)

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and has since been reported in Myanmar (Thaung 2005) and South Africa (Doidge 1950). This rust has pycnidia, aecidia, and uredosori that are unknown in the type species of the genus (Sydow & Petrak 1928, Cummins 1937). The present paper reports the occurrence of *Masseella flueggeae* on *Flueggea virosa* for the first time in Pakistan. In addition, a noteworthy rust, *Pucciniastrum pyrolae* on *Pyrola rotundifolia* subsp. *karakoramica*, is reported here as a new host record in Pakistan.

## Materials and methods

Rusted specimens were collected in Azad Kashmir, Neelum Valley, Lawat and northern areas, Fairy Meadows, Pakistan. Freehand sections of infected tissues and spores were mounted in lactophenol and gently heated to a boiling point. The preparations were observed under a NIKON YS 100 microscope. Spores and sori were drawn using a camera lucida (Ernst Leitz Wetzlar, Germany). Spores were measured with an ocular micrometer. At least 25 spores were measured for each spore state. The specimens were deposited in the Herbarium of the Botany Department, University of the Punjab, Lahore (LAH).

## Recorded species

*Masseella flueggeae* Syd., Ann. Mycol. 26: 424 (1928).

FIG. 1

MATERIAL EXAMINED: Pakistan, Azad Kashmir, Neelum valley, Lawat, on *Flueggea virosa* (Willd.) Voigt (*Euphorbiaceae*), 16 Aug 2009, Abdul Nasir Khalid 130 (LAH 1130).

SPERMOGONIA and AECIA unknown. UREDINIA amphigenous, forming groups, yellow to yellowish orange, subepidermal, mostly intermixed with telia. UREDINIOSPORES ellipsoid to obovoid, hyaline to yellow,  $15\text{--}19 \times 18\text{--}27 \mu\text{m}$ , wall  $1.5\text{--}2 \mu\text{m}$  thick, echinulate to verrucose, germ pores obscure. TELIA amphigenous, crowded, mostly along veins or margins of leaf, causing malformations, subepidermal, arising in uredosori, becoming erumpent as hair-like columns, orange to yellowish brown or chestnut brown. TELIOSPORES one-celled, sessile with hyphal attachment organs resembling pedicels, up to  $34 \mu\text{m}$  long, ellipsoid to broadly ellipsoid or cylindric to angular,  $16\text{--}24 \times 23\text{--}47 \mu\text{m}$ , embedded in mucilaginous mass, germ pore apical, wall striate, yellowish brown to chestnut brown,  $4\text{--}6 \mu\text{m}$  thick at sides and  $4\text{--}7 \mu\text{m}$  thick apically.

*Pucciniastrum pyrolae* Arthur, North Amer. Fl. 7: 108 (1907).

FIG. 2

MATERIAL EXAMINED: Pakistan, Northern Areas of Pakistan, Fairy Meadows, Bial Camp, at 3,036 m a.s.l., On *Pyrola rotundifolia* subsp. *karakoramica* (Křisa) Y.J. Nasir (*Ericaceae*), with II stage, 11 Aug 2007. Najam-ul-Sehar Afshan G07 (LAH NSA 1119).

SPERMOGONIA, AECIA, and TELIA unknown. UREDINIA hypophyllous, covered by epidermis, yellowish orange, rounded, minute, in form of group, covered

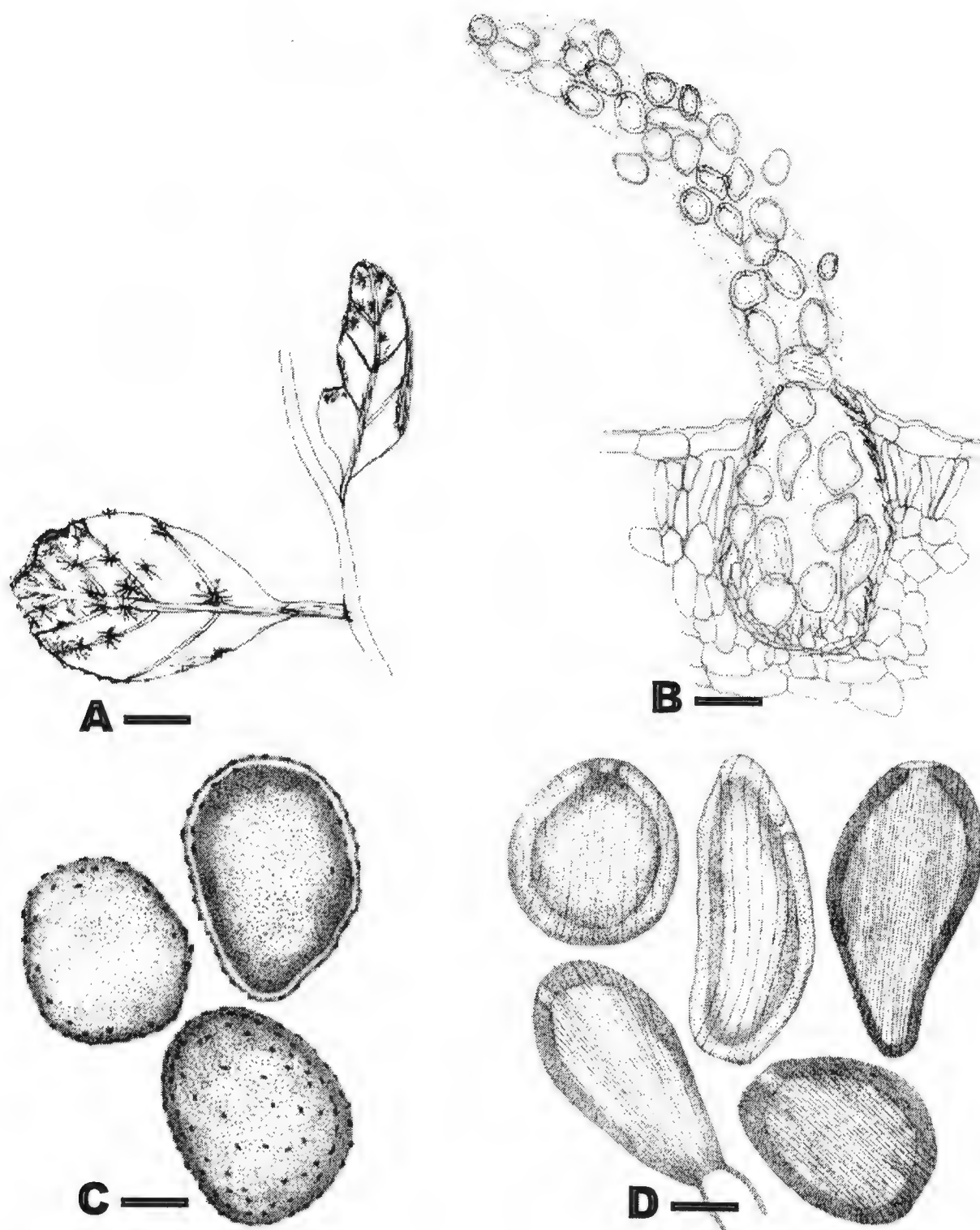


FIG. 1. *Masseella flueggeae*.

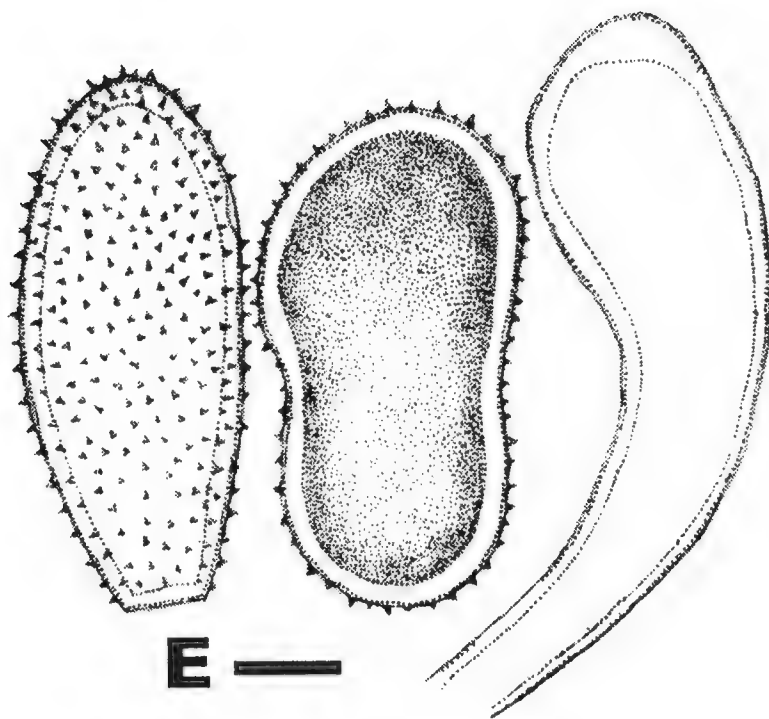
A. Drawing of host plant showing infected parts.

B. A telial sorus showing the development of spore column and mucilage-secreting hyphae.

C. Urediniospores with echinulate to verrucose wall ornamentation.

D. Mature teliospores with striate walls.

Scale bars = 10  $\mu$ m.

FIG. 2. *Pucciniastrum pyrolae*.

Drawings of mature urediniospores and apex of a paraphysis. Scale bar = 8  $\mu\text{m}$ .

by a peridium of hyphal cells, releasing spores by an ostiolar opening, 0.1–0.2  $\times$  0.2–0.4 mm. UREDINIOSPORES ovoid to obovoid or ellipsoid, 13–18  $\times$  26–37  $\mu\text{m}$  (mean = 16.0  $\times$  32.0  $\mu\text{m}$ ); wall 1.8–3  $\mu\text{m}$  thick, hyaline to yellow, echinulate; germ pores obscure. Paraphyses clavate, hyaline or yellowish, 13–15  $\times$  47–71  $\mu\text{m}$ .

*Pucciniastrum pyrolae* has previously been reported on leaves of *Pyrola secunda* L. from Fairy Meadows and Gilgit by Kaneko (1993). *Pyrola rotundifolia* subsp. *karakoramica* is a new host for this rust fungus in Pakistan.

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Two new species of *Stachybotrys* from soil

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**Abstract** — Two new species are described and illustrated: *Stachybotrys jiangziensis* and *S. xigazenensis*, both from soil in China. The type specimens (dried cultures) and living cultures are deposited in the Herbarium of Shandong Agricultural University Plant Pathology (HSAUP). Isotypes are kept in the Herbarium of Institute of Microbiology, Academia Sinica (HMAS).

**Key words** — taxonomy, soil fungi, dematiaceous hyphomycetes

## Introduction

*Stachybotrys* Corda was erected in 1837, and since then 96 epithets have been proposed in the genus (Index Fungorum 2010). This genus is characterized by distinct, mononematous conidiophores bearing an apical cluster of several swollen phialides producing unicellular phialoconidia that become aggregated in globose masses. In the course of a survey of soil dematiaceous hyphomycetes in China, several unusual species of *Stachybotrys* were collected. Two of them are described as new species, *S. jiangziensis* and *S. xigazenensis*.

## Taxonomy

*Stachybotrys jiangziensis* Y.M. Wu & T.Y. Zhang, sp. nov.

FIG. 1

MYCOBANK MB 518786

*Coloniae in CMA effusae, atrogriseae vel nigrae. Hyphis ramosis, septatis, laevibus, hyalinis vel subhyalinis, 1.5–3 µm crassis. Conidiophora erecta, 2–4-septata, basim versus subhyalina, supra griseo-brunnea, levia, 60–80 µm longa, ad basim 4–5 µm diam. Phialides 6–8 ad apicem conidiophori productae, pallide brunneae, leves, 8–10 × 5–7 µm. Conidia tuberculata, globosa vel subglobosa, brunnea vel atrobrunnea, 6–9 µm diam.*

**HOLOTYPE:** China. Tibet, Jiangzi, from a grassland soil, altitude 4050 m, 9 Sept. 2007, Y.M. Wu, HSAUPII<sub>07</sub>0881, **holotype**; HMAS 196256, **isotype**.

**ETYMOLOGY:** in reference to the type locality.

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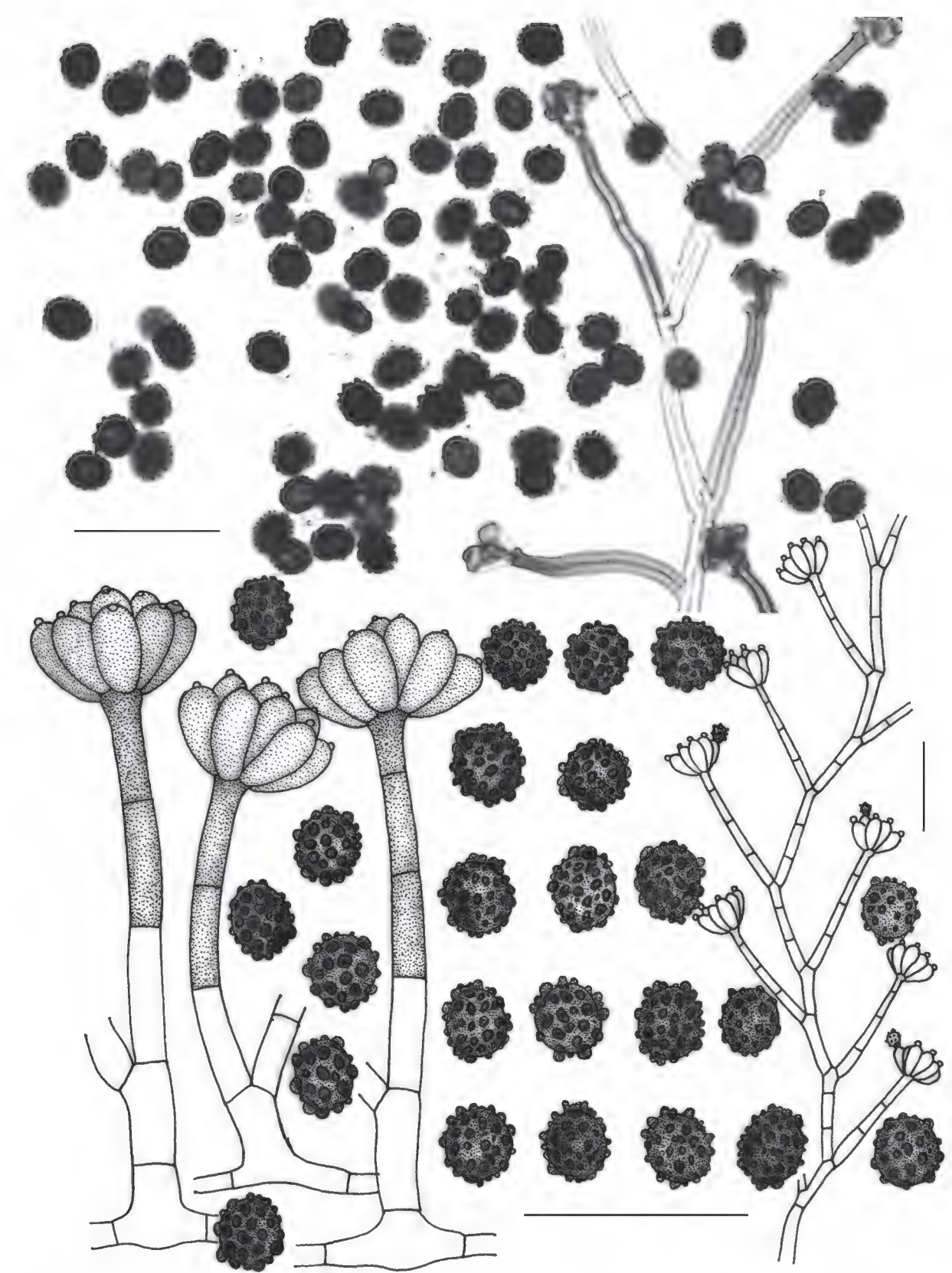


FIG. 1. *Stachybotrys jiangziensis* (ex holotype). Conidia, conidiophores, and conidiogenous cells. Above: photomicrographs. Below: drawings. (Bars = 25  $\mu$ m).

Colonies on CMA (cornmeal agar) at 25°C for 21 days 4–6 cm diam., effuse, darkish grey to black. Mycelium mostly superficial, partly immersed. Hyphae branched, septate, smooth, hyaline to subhyaline, 1.5–3  $\mu$ m wide. Conidiophores

erect, branched, 2–4-septate, subhyaline near the base, greyish brown above, smooth, 60–80  $\mu\text{m}$  long, 4–5  $\mu\text{m}$  wide near base. Phialides borne in groups of 6–8 at the apices of conidiophores, pale brown, smooth,  $8\text{--}10 \times 5\text{--}7 \mu\text{m}$ . Conidia globose to subglobose, tuberculate, brown to dark brown, 6–9  $\mu\text{m}$  in diameter.

In conidial morphology this fungus somewhat resembles *Stachybotrys nilagirica* Subram. (Subramanian 1957) and *S. sphaerospora* Morgan-Jones & R.C. Sinclair (Morgan-Jones & Sinclair 1980). However *S. nilagirica* has larger conidia (16–20  $\mu\text{m}$  diam.) and *S. sphaerospora* larger (11–12  $\mu\text{m}$  diam.), ridged conidia than *S. jiangziensis*.

***Stachybotrys xigazensis* Y.M. Wu & T.Y. Zhang, sp. nov.**

FIG. 2

MYCOBANK MB 518787

*Coloniae in CMA effusae, atrogriseae vel nigrae. Hyphis ramosis, septatis, laevibus, hyalinis vel subhyalinis, 2–3  $\mu\text{m}$  crassis. Conidiophora erecta, 1–4-septata, basim versus subhyalina, supra griseo-brunnea, verrucosa, interdum granulis magnis tecta, 60–100  $\mu\text{m}$  longa, ad basim 4–6  $\mu\text{m}$  diam. Phialides 6–8 ad apicem conidiophori productae, pallide brunneae, leves,  $7\text{--}10 \times 5\text{--}8 \mu\text{m}$ . Conidia ovoidea, ellipsoidea vel oblonga, tuberculata, brunnea vel atrobrunnea,  $9\text{--}12.5 \times 7.5\text{--}10 \mu\text{m}$ .*



FIG. 2. *Stachybotrys xigazensis* (ex holotype). Conidia, conidiophores, and conidiogenous cells. Left: photomicrographs. Right: drawings. (Bars = 25  $\mu\text{m}$ ).

HOLOTYPE: China. Tibet, Xigazen, from a grassland soil, altitude 4150 m, 19 Sept. 2007, Y.M. Wu, HSAUPII<sub>07</sub>1450, **holotype**; HMAS 196257, **isotype**.

ETYMOLOGY: in reference to the type locality.

Colonies on CMA at 25°C for 21 days 5–8 cm diam., effuse, darkish grey to black. Mycelium mostly superficial, partly immersed. Hyphae branched, septate, smooth, hyaline to subhyaline, 2–3 µm wide. Conidiophores erect, sympodially branched, 1–4-septate, subhyaline near the base, greyish brown above, verrucose, sometime covered with large granules, 60–100 µm long, 4–6 µm wide near base. Phialides borne in groups of 6–8 at the apices of conidiophores, pale brown, smooth, 7–10 × 5–8 µm. Conidia ovoid, ellipsoid or oblong, tuberculate, brown to dark brown, 9–12.5 × 7.5–10 µm.

This fungus somewhat resembles *Stachybotrys chartarum* (Ehrenb.) S. Hughes (Hughes 1958) and *S. microspora* (B.L. Mathur & Sankhla) S.C. Jong & E.E. Davis (Jong & Davis 1976) in conidial colour and size, but *S. xigazensis* has more obviously tuberculate conidia. In addition, the conidia of *S. xigazensis* are larger than those of *S. microspora* (6–8 × 4–5 µm) and wider than those of *S. chartarum* (7–12 × 4–6 µm).

### Acknowledgments

The authors are grateful for pre-submission comments and suggestions provided by Dr. Eric McKenzie, Prof. Y.L. Guo, and Dr. Shaun Pennycook. This project was supported by the National Science Foundation of China (no. 30670014 & 30499340).

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## MYCOTAXON

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## The genus *Placidiopsis* in the Iberian Peninsula and the Balearic Islands

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**Abstract** — A taxonomic revision of the genus *Placidiopsis* in the Iberian Peninsula and the Balearic Islands is provided. A total of 500 specimens were studied. A detailed description of the morphology, anatomy, ecology, and distributional rank is presented for each species. Additionally, a key to *Placidiopsis* species is included. The genus is represented by four species in the studied region, with *P. cavicola* and *P. cinereoides* known only from the type localities and *P. cinerascens* and *P. custnani* common in the eastern half of the region. These data expand considerably the ecological and distributional range of these species in the Iberian Peninsula.

**Key words** — catapyrenioid lichens, distribution, Spain, Portugal, *Verrucariaceae*

### Introduction

*Placidiopsis* Beltr. (*Verrucariaceae*) is a genus of squamulose lichens closely related to *Catapyrenium* s. str., although recent phylogenetic analyses (Gueidan et al. 2007, 2009; Prieto et al. 2010) concluded that both genera were different entities. The two genera are morphologically differentiated by uniseptate ascospores in *Placidiopsis* and simple ascospores in *Catapyrenium* s. str. *Placidiopsis* species are characterized by squamulose thalli attached to the substrate by a rhizohyphal web, a central bundle of rhizohyphae, or rhizines. The upper cortex is either absent or *cinereum*-type (Breuss 1996, 2002; Prieto et al. 2010), the photobiont is a chlorococcoid alga (Breuss 2002), the medulla is proso- or subparaplechtenchymatous, and the lower cortex is (sub)paraplectenchymatous when present. Perithecia are immersed, with or without an apical involucrellum, asci are clavate with an ocular chamber, and pycnidia have never been observed (Breuss 2002).

Members of the group inhabit arid, semiarid, and arctic–alpine regions in the Northern Hemisphere (Breuss 2002). Ecological preferences of the genus

include soil, rock, detritus, or bryophytes occurring in calciferous or acid substrates. *Placidiopsis* comprises 12 species world-wide (Breuss 2010), many of which appear rare and restricted either to their type localities (e.g. *P. cavicola*, *P. cervinula* (Nyl.) Vain., *P. cinereoides*) or to very small areas (e.g. *P. hamadicola* Bredkina, *P. tirolensis* Breuss).

Breuss (1996), who until now has published the only complete treatment of the genus, reported few specimens for the Iberian Peninsula. Therefore exhaustive collection and more in-depth research of *Placidiopsis* species was necessary in order to establish the true extent of the genus in the Iberian Peninsula and the Balearic Islands. The current research is part of the project Spanish Lichenological Flora.

Materials & methods

This study is mainly based on material collected by the authors in the Iberian Peninsula and the Balearic Islands during 2005–2009. The specimens are deposited in MA herbarium. In addition, collections in Iberia (BCC, BCN, LEB, LISU, MA, MACB, MAF, SANT, VAL, VIT), other European and North American herbaria (ABL, ARIZ, ASU, B, BM, COLO, GB, H, HAL, L, LI, NY, PRM, S, TUR), and personal herbaria (C. Keller, G. Aragón) were revised. Approximately 500 specimens in total were studied.

Observations and measurements were made using a Nikon SMZ–800 dissecting microscope and an Olympus BX 51 microscope. Thallus cross-sections (14–20 µm thick) were made with a Leica CM 1850 UV freezing microtome. Sections were observed and measured in water or occasionally lactophenol cotton blue. For anatomical studies, ten specimens per species were analysed (when available), and ten measurements of each specimen on different squamules were carried out. The limited material of some species and the poor condition of others led to a lower number of measurements in some cases. Measurements are expressed as the mean ± standard deviation (SD) with the extremes within parentheses; length/wide ratios (l/w) were calculated for ascospores. Distributional maps were drawn with ArcView GIS 3.1, based on UTM coordinates (WGS84 Datum).

For each taxon, we cite the basionym, type specimen, and type location, but not previously published synonyms (see Breuss 1996).

Results

Of the four *Placidiopsis* species found in the Iberian Peninsula and the Balearic Islands, two — *P. cavicola*, *P. cinereoides* — are known only from their type localities and two — *P. cinerascens*, *P. custnani* — are more common than previously believed and found throughout the studied area.

Key to the known *Placidiopsis* species in the Iberian Peninsula

- 1. On rocks, squamules up to 0.5 mm .....*P. cavicola*
- 1. On soil, squamules up to 2–3 mm .....2

- 2. Rhizohyphae colourless ..... *P. cinerascens*
- 2. Rhizohyphae dark ..... 3
- 3. Squamules with down-rolled margins, rhizohyphae attached in a central holdfast  
(looking like a rhizine), ascospores  $15\text{--}22 \times 5\text{--}7 \mu\text{m}$  ..... *P. custnani*
- 3. Squamules without downrolled margins, central holdfast absent,  
ascospores  $22\text{--}28 \times 7\text{--}8 \mu\text{m}$  ..... *P. cinereoides*

***Placidiopsis cavicola*** Etayo & Breuss, Österr. Z. Pilzk. 3: 21 (1994) FIGS. 1A, 2A

[TYPE: Spain, Navarra, Larra, Isaba, Añelarra, cave A-50, 5 m depth, on calcareous flagstone, 2154 m, J. Etayo & J.I. Calvo, 19/08/1992 (Herb. Etayo, HOLOTYPE; LI 271012, ISOTYPE!).]

**MORPHOLOGY**—Thallus squamulose, composed of very small squamules,  $\leq 0.5$  mm broad, flat, crenulate, adjacent to slightly overlapping. Upper surface green to light brown; lower surface brown, with colourless to brown rhizohyphae.

**ANATOMY**—Thallus  $100\text{--}150(-250) \mu\text{m}$  thick, upper cortex  $10\text{--}20 \mu\text{m}$  thick, with cells of  $4\text{--}6 \mu\text{m}$  diam, epinecral layer lacking. Algal layer distributed over nearly the entire thallus, with algal cells of  $5\text{--}9 \mu\text{m}$ ; lower cortex not clearly delimited. Rhizohyphae colourless to brownish, ca.  $4 \mu\text{m}$  thick.

Perithecia  $150\text{--}250 \mu\text{m}$  wide, with a colourless exciple. Asci clavate,  $45\text{--}55 \times 15\text{--}20 \mu\text{m}$ , ascospores septate,  $13\text{--}17 \times 6\text{--}7 \mu\text{m}$  (Etayo & Breuss 1994). Pycnidia absent.

**ECOLOGY & DISTRIBUTION** — *Placidiopsis cavicola* was collected on rock, growing over a thin algal or debris layer in a calcareous cave in the subalpine belt of the Pyrenees, over  $2100\text{--}2200$  m altitude (Etayo & Breuss 1994).

The species is known only from the type locality in Navarra, Spain; it may be more widely distributed, however, as it has probably been overlooked due to its small size.

**COMMENTS**—*Placidiopsis cavicola* resembles *P. minor* R.C. Harris in that both species have small squamules (no more than 1 mm) and grow on rocks. However, *P. cavicola* has crenulated and non-pruinose squamules, while *P. minor* has roundish to slightly lobed and pruinose squamules; moreover, the spores are bigger in *P. cavicola* ( $8\text{--}10 \times 4\text{--}5 \mu\text{m}$  in *P. minor*). *Placidiopsis minor* has not been found until now in the Iberian Peninsula and has previously been known only from North America and Greenland (Breuss 1996).

***Placidiopsis cinerascens*** (Nyl.) Breuss, Plant Syst. Evol. 148: 315 (1985) FIGS. 1B, 2A

[TYPE: Gallia merid., Beaucaire, W. Nylander (H-NYL 4021, HOLOTYPE!).]

= *Placidiopsis tenella* (Nyl.) Zahlbr., Catal. Lich. Univ. 1: 240 (1921)

[TYPE: Oran, Balansa (H-NYL 3944!, LECTOTYPE, designated by Cl. Roux in herbarium).]

**MORPHOLOGY**— Thallus squamulose, squamules up to 3 mm wide, scattered to contiguous, flattened to slightly convex, rounded to lobed or crenate. Upper surface whitish, greenish grey or brownish grey, pruinose or not; lower surface pale with colourless rhizohyphae.

**ANATOMY**— Thallus (110–)  $226 \pm 48.9$  (–320)  $\mu\text{m}$  thick, with or without epinecral layer, up to 50  $\mu\text{m}$  when present; upper cortex (5–)  $19.1 \pm 8.1$  (–37.5)  $\mu\text{m}$  thick, paraplectenchymatous, with roundish-subangular cells of (4–)  $7.1 \pm 1.4$  (–11)  $\mu\text{m}$  diam. Algal layer distributed over almost the entire thallus, 50–175  $\mu\text{m}$  thick, with cells (3–)  $6.5 \pm 1.7$  (–12)  $\mu\text{m}$  diam. Medulla not clearly delimited from the algal layer, composed of globular cells (4–)  $7.5 \pm 1.5$  (–11)  $\mu\text{m}$  diam; lower cortex lacking. Rhizohyphae colourless, (2.5–)  $3.2 \pm 0.4$  (–4)  $\mu\text{m}$ .

Perithecia slightly pyriform to globose, up to 300  $\mu\text{m}$  wide, exciple hyaline to brownish, up to ca. 30  $\mu\text{m}$  thick, darker in the ostiole, with or without a small apical involucrellum. Asci clavate, 55–65  $\times$  11–16  $\mu\text{m}$  (Breuss 1996), with a small ocular chamber; ascospores biseriate, hyaline, septate (occasionally simple), (12–)  $16.4 \pm 1.9$  (–21)  $\times$  (5–)  $6.2 \pm 0.5$  (–7)  $\mu\text{m}$ , l/w ratio (1.7–)  $2.6 \pm 0.4$  (–3.3). Pycnidia absent.

**ECOLOGY & DISTRIBUTION**—The species shows preferences for soil and rock ledges on calcareous and gypsiferous substrates. It was found in shrublands with *Buxus sempervirens* L., *Lavandula latifolia* Medik., *Lycium* sp., *Rosmarinus officinalis* L., and *Thymus* sp. in dry and open habitats, but also collected in *Pinus halepensis* Mill., *Juniperus thurifera* L. and *Quercus ilex* subsp. *ballota* (Desf.) Samp. forests. *Placidiopsis cinerascens* has been frequently found together with *Anthracoarpon virescens* (Zahlbr.) Breuss, *Endocarpon pusillum* Hedw., *Placidiopsis custnani*, or *Placidium pilosellum* (Breuss) Breuss. In the studied area, *P. cinerascens* was found between the sea level and 1300(–1800) m altitude.

Until now, *P. cinerascens* was little collected in the Iberian Peninsula and recorded from only 5 southern and eastern provinces of Spain although also known from Portugal (Barreno et al. 1989, Breuss 1996, Etayo & Breuss 1996). There are few records of *P. cinerascens* reported as *P. tenella* in Spain (Boom & Gómez-Bolea 1991, Etayo 1992, Gutierrez & Casares 1994, Guerra et al. 1995); as these specimens could not be examined, their data are not included in the maps.

Our data indicate that *P. cinerascens*, relatively abundant in the Iberian Peninsula, is more common than previously thought. New data extend the known distribution of the species in the Iberian Peninsula, mainly from central, southern and southeastern Spain, with many collections constituting first provincial records. Although present throughout the Iberian Peninsula with



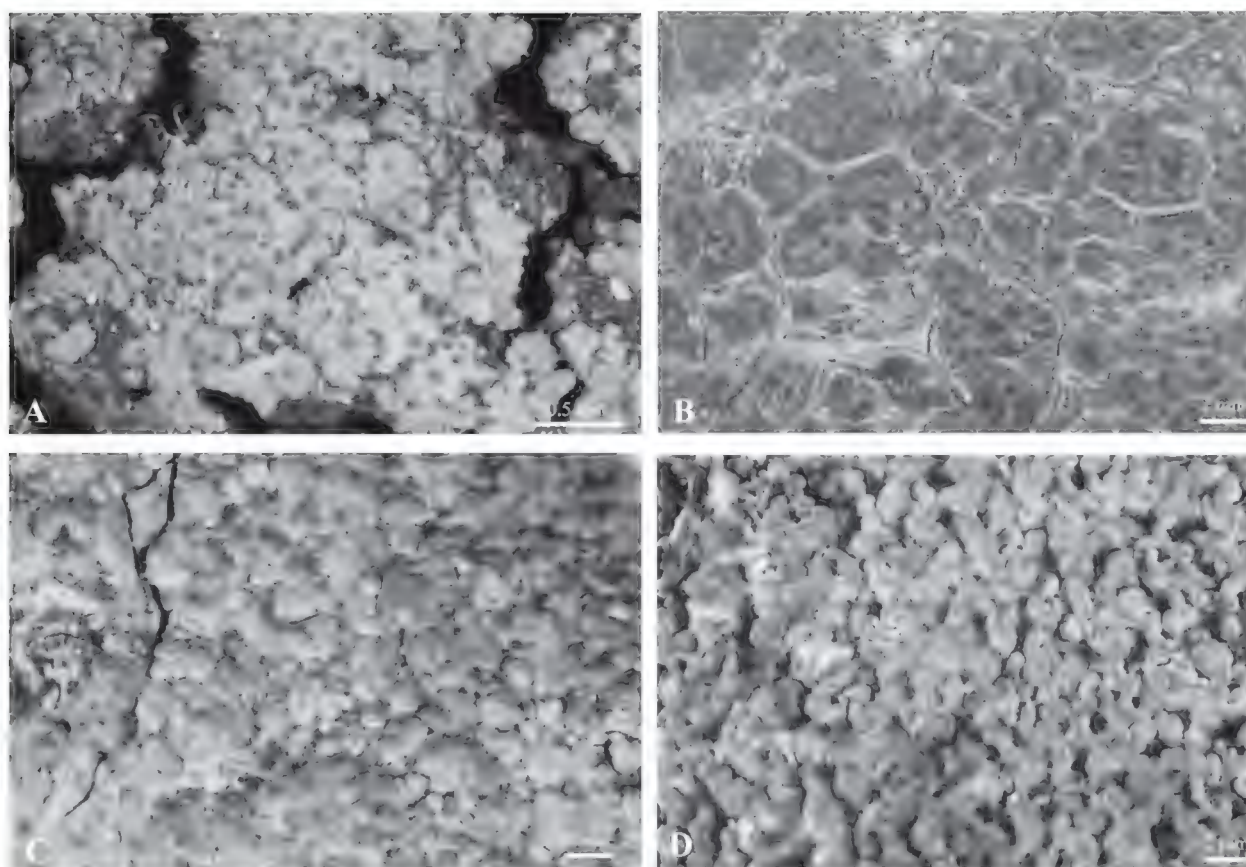


FIGURE 1. Habit of species of *Placidiopsis*.  
A, *P. cavicola*; B, *P. cinerascens*; C, *P. cinereoides*; D, *P. custnani*.

some occurrence in the west, the species is especially prevalent in the eastern half; it is relatively common in the Balearic Islands.

*Placidiopsis cinerascens* is widely distributed in Mediterranean and arid climates throughout the European mediterranean region as well as in central Asia, Mexico, northern Africa, and SW North America (Breuss 2002).

COMMENTS— *Placidiopsis cinerascens* was synonymized with *P. tenella* based on morphological and genetic similarities (Prieto et al. 2010). The presence of an involucrellum in *P. tenella* is not a valid character state, because it does not appear in all ascomata within the same specimen or even in the same squamule. Therefore, *Placidiopsis tenella* cannot be distinguished from *P. cinerascens* using this character.

REPRESENTATIVE SPECIMENS — SPAIN. ALBACETE: Riópar, Calar del Mundo, subida por la Fuente de las Raigadas, 549451 E, 4256284 N, 1320 m, G. Aragón, R. Belinchón & M. Prieto, 31/01/2007, M. Prieto 664b. ALICANTE: Orihuela, 680582 E, 4218299 N, suelos calizos, 35 m, M. Prieto, 11/04/2006, M. Prieto 589b. ALMERÍA: Turrillas, Sierra Alhamilla, 565453 E, 4098680 N, 1300 m, suelos calizos, I. Martínez, M.A.G. Otálora & M. Prieto, 29/11/2005, M. Prieto 515. BARCELONA: Can Grau, Sierra del Garraf, 403256 E, 4573874 N, 279 m, M. Prieto, 08/07/2004, M. Prieto 1654. BURGOS: Oña, carretera hacia Villanueva de los Montes, Sierra de Tesla, 466595 E, 4733638 N, repisas calizas, 590 m, I. Martínez & M. Prieto, 23/08/2007, M. Prieto 1166. CÁCERES: Torrejón El Rubio, castillo de Monfragüe, 244525 E, 4414075 N, sobre mortero de un muro,



661 m, M. Prieto, 14/01/2007, M. Prieto 662 (MA 16302). **CÁDIZ**: Grazalema, Sierra de Grazalema, carretera hacia Zahara de la Sierra, antes del Puerto de los Acebuches, 287604 E, 4075692 N, 870 m, oquedades de rocas calizas, R. Belinchón, I. Martínez & M. Prieto, 13/06/2008, M. Prieto 1474. **CASTELLÓN**: Cabanes, dessert de les Palmes, 248364 E, 4448733 N, fisuras calizas, 290 m, M. Prieto, 14/03/2008, M. Prieto 1410, 1411 (MA 16302). **CUENCA**: Poyatos, 582121 E, 4474336 N; repisas de rocas calizas en pinar, 1046 m, M. Prieto, 05/04/2007, M. Prieto 942. **GRANADA**: Sierra Nevada, antes de Prado Llano, 460330 E, 4107723 N, suelo calizo, 1880 m, M. Prieto, 26/06/2008, M. Prieto 1523. **LA RIOJA**: Foncea, 497345 E, 4718904 N, suelo entre rocas calizas, 860 m, I. Martínez & M. Prieto, 27/08/2007, M. Prieto 1154, 1160. **LEÓN**: Miñera de Luna, 267275 E, 4751065 N, suelos calizos en sabinar, 1130 m, M. Prieto, 18/05/2006, M. Prieto 617. **LÉRIDA**: Alfés, Timoneda, aeròdrom d'Alfés, 30TCG00, terrícola, 240 m, J. Perez-Redondo, 12/01/1992, BCC 12680. **MADRID**: Patones de Arriba, 459550 E, 4524950 N, 834 m, M. Prieto, 01/05/2008, M. Prieto 1520. **MÁLAGA**: Parauta, Sierra de las Nieves, estribaciones del pinsapar de cerro Alcojona, cerca del pinsapo de la Escalereta, 318103 E, 4060026 N, 1164 m, R. Belinchón, I. Martínez & M. Prieto, 12/06/2008, M. Prieto 1454b. **MALLORCA**: Caimari, Sierra de Tramuntana, 681883 E, 4759848 N, fisuras calizas, 500 m, M. Prieto, 15/04/2007, M. Prieto 904 (MA 16304). **NAVARRA**: Rada, Bárdenas Reales, 616320 E, 4686664 N, suelo calizo, I. Martínez & M. Prieto, 22/08/2007, M. Prieto 1131. **PALENCIA**: Piedrasluengas, Puerto de Piedrasluengas, 381275 N, 4767675 E, fisuras calizas, 1355 m, G. Aragón, A. García & M. Prieto, 21/07/2005, M. Prieto 108 (MA 16397). **TOLEDO**: carretera hacia Villacañas, 476725 E, 4378425 N, M. Prieto, 21/01/2007, M. Prieto 657. **VALENCIA**: carretera de Utiel a Estenas, Sierra de Juan Navarro, 659189 N, 4384368 E, suelos calizos en coscojar, 892 m, M. Prieto, 22/02/2008, M. Prieto 1328, 1330. **ZARAGOZA**: Calcena, 606269 E, 4610745 N, repisas calizas, 890 m, I. Martínez & M. Prieto, 21/08/2007, M. Prieto 1116. **PORTUGAL**: Alvados, Serra de Aire e os Candeiros, grutas, 521231 E, 4376584 N, suelos calizos, 445 m, M.A.G. Otálora & M. Prieto, 27/09/2007, M. Prieto 1257 (MA 16309), 1263, 1266.

*Placidiopsis cinereoides* Breuss, Österr. Z. Pilzk. 5: 84 (1996)

FIGS. 1C, 2B

[TYPE: España, Palencia, Pico Curavacas, sobre conglomerado silíceo, 1900–2100 m, M.E. López de Silanes, 09/09/1990 (SANT 7072, HOLOTYPE!; LI 271013, ISOTYPE!).]

**MORPHOLOGY**— Thallus squamulose, composed of contiguous to slightly overlapping squamules, forming a compact rosette; squamules finely lobulate to crenate, flat to slightly convex, up to 2 mm wide; upper surface whitish to greenish grey-brown; lower surface dark with brown rhizohyphae.

**ANATOMY**— Thallus 200–400 µm thick; upper cortex up to 20 µm thick, with roundish-subangular cells of 5–8 µm diam; with or without epinecral layer, up to 50 µm when present. Algal layer filling almost half of the thallus, with cells (3)  $5 \pm 1.3$  (6) µm diam. Medulla subparaplectenchymatous with globular cells of 8–11 µm diam, brownish in the lower zone; lower cortex paraplectenchymatous, of more densely aggregated cells. Rhizohyphae brown, ca. 4 µm.

Perithecia globose, 200–400 µm wide, exciple colourless to brownish; asci 65–80 × 16–20 µm (Breuss 1996), ascospores biseriate, hyaline, septate (occasionally simple), (20) 22–28 (30) × (6.5) 7–8 (8.5) µm. Pycnidia absent.

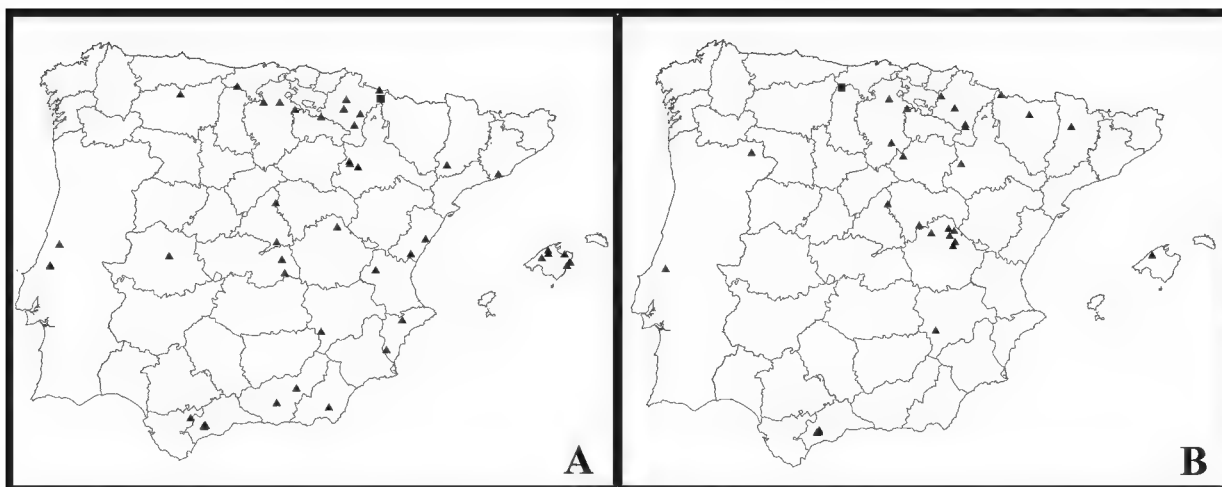


FIGURE 2. Distribution of *Placidiopsis* species in the Iberian Peninsula and the Balearic Islands. A, *P. cinerascens* (▲) and *P. cavicola* (■); B, *P. custnani* (▲) and *P. cinereoides* (■).

**ECOLOGY & DISTRIBUTION**—The species was found growing in a cave over siliceous substrate, in the northern slope, at 1900–2100 m altitude. *Placidiopsis cinereoides* is known only from the type locality in the north half of the Iberian Peninsula.

**COMMENTS**—The species is well recognized by the presence of larger ascospores and the rosette-like growth.

***Placidiopsis custnani*** (A. Massal.) Körb., *Parerga Lichenol.* 305 (1863) FIGS. 1D, 2B

[TYPE: in opp. Scorgano (Custnano), Verona, A. Massalongo. (A. Massal., *Lich. exs. Ital.* 187, M LECTOTYPE, GE, L, M, W! ISOLECTOTYPES).]

**MORPHOLOGY**—Thallus squamulose, composed of scattered to contiguous squamules; squamules lobulate to crenate, up to 2(–3) mm wide, with margins ascending and down-rolled; upper surface olive green to brownish or greyish, pruinose or not; lower surface dark brown to black or carbonaceous but pale at margins; attached by a central holdfast of dark rhizohyphae, forming a rhizine-like structure.

**ANATOMY**—Thallus (180)  $262.1 \pm 51.7$  (380)  $\mu\text{m}$  thick, upper cortex (7.5)  $25.1 \pm 9.6$  (50)  $\mu\text{m}$  thick, paraplectenchymatous, with roundish-subangular cells of (3)  $6.4 \pm 1.7$  (10)  $\mu\text{m}$  diam; with or without epinecral layer, up to 50  $\mu\text{m}$  when present. Algal layer 55–175  $\mu\text{m}$  thick, with cells (5)  $7.1 \pm 1.2$  (11)  $\mu\text{m}$  diam. Medulla (37.5)  $89.5 \pm 31.2$  (150)  $\mu\text{m}$ , composed mainly of globular cells, (4)  $6.7 \pm 1.3$  (10)  $\mu\text{m}$  diam; lower cortex not clearly delimited. Rhizohyphae colourless, (3)  $3.5 \pm 0.5$  (4)  $\mu\text{m}$ .

Perithecia pyriform to globose, up to ca. 200  $\mu\text{m}$  wide, exciple hyaline to brown or black, darker on the ostiole, up to ca. 20  $\mu\text{m}$  thick; asci clavate, 50–70  $\times$  10–14  $\mu\text{m}$  (Breuss 1996), with a small ocular chamber; ascospores biseriate,

hyaline, septate, (15)  $18.2 \pm 1.6$  (22)  $\times$  (5)  $6.1 \pm 0.5$  (7.2)  $\mu\text{m}$ , l/w ratio (2.3)  $3 \pm 0.3$  (3.8). Pycnidia absent.

**ECOLOGY & DISTRIBUTION** — *Placidiopsis custnani* shows preferences for calcareous soils. We have found it mainly in *Pinus halepensis*, *Juniperus thurifera*, and *Quercus ilex* subsp. *ballota* forests; it was found together with *Placidium pilosellum* and sometimes with *Placidiopsis cinerascens*, usually mixed with bryophytes. In the studied area, *P. custnani* was found between 300 and ca. 1500 m altitude.

*Placidiopsis custnani* has been very infrequently recorded in the Iberian Peninsula. Paz-Bermúdez et al. (2009) reported the second record of the species in the studied region, previously cited from Mallorca (Breuss 1996); this specimen constituted the first record from Portugal. Nevertheless, there are two more records in Spain from 1994 (Hladun & Llimona 2002–07).

Our data considerably extend the known distribution of *P. custnani* in the Iberian Peninsula and the Balearic Islands, with many of the collections constituting first provincial records. The species has been found mainly in central and northern Spain, although there are some localities in southern Spain. In general, the species inhabits colder places than *P. cinerascens*.

Worldwide distribution of *Placidiopsis custnani* includes central Europe reaching northern Europe and the Mediterranean Region (Breuss 1996).

**COMMENTS**— *Placidiopsis custnani* is easily identified by the presence of ascending squamules with down-rolled margins.

**REPRESENTATIVE SPECIMENS** — **SPAIN. ALBACETE:** Riópar, Sierra de Alcaraz, Calar del Mundo, 555692 E, 426654N, suelo y fisuras calizas, 1530 m, G. Aragón, R. Belinchón y M. Prieto, 01/02/2007, M. Prieto 672, 674. **BURGOS:** Contreras, pista hacia Santo Domingo de Silos, Sabinas del Arlanza, 465731 E, 4648768 N, 1276 m, suelo entre musgos, I. Martínez & M. Prieto, 23/08/2007, M. Prieto 1190. Panizares, Sierra de Tesla, 461124 E, 4738773 N, 641 m, suelo entre matorral con boj, I. Martínez & M. Prieto, 23/08/2007, M. Prieto 1168, 1169. **CUENCA:** Las Majadas, Los Callejones, 584688 E, 4459765 N, suelo limoso, 1410 m, M. Prieto, 05/04/2007, M. Prieto 964, 980. **GUADALAJARA:** Sacedón, carretera hacia Auñón, embalse de Buendía, 521366 E, 4481909 N, 752 m, suelos calizos, M. Prieto, 31/03/2007, M. Prieto 790. **HUESCA:** Laguarda, carretera hacia Sabiñánigo, 746634 E, 4706241 N, suelos calizos, 600-700 m, M. Prieto, 04/03/2007, M. Prieto 709 (MA 16303). **LA RIOJA:** Foncea, 497345 E, 4718904 N, suelos calizos entre matorral con boj, sabina y encinas, 860 m, I. Martínez & M. Prieto, 23/08/2007, M. Prieto 1151 (MA 16310). **LÉRIDA:** Abella de la Conca, Sierra de Carreu, camí Herba-Savina, 832233 E, 4681537 N, suelo entre encinar, 831 m, M. Prieto, 12/08/2008, M. Prieto 1590. **MADRID:** Patones de Arriba, 459550 E, 4524950 N, suelos calizos, 834 m, M. Prieto, 01/05/2008, M. Prieto 1521. **MÁLAGA:** Parauta, Sierra de las Nieves, estribaciones del pinsapar de cerro Alcojona, cerca del pinsapo de la Escalereta, 318103 E, 4060026 N, repisa caliza, 1164 m, I. Martínez & M. Prieto, 12/06/2008, M. Prieto 1452. **MALLORCA:** umgebung von Soller, Hohe im Ort, betretener Boden, C. & J. Poelt, 07/04/1964, M. **NAVARRA:** Bárdenas Reales, hacia el embalse de El Ferial, 616227 E, 4681607 N, suelos yesíferos,

*Juniperus phoenicea* y *Quercus coccifera*, 362 m, I. Martínez & M. Prieto, 22/08/2007, M. Prieto 1128. **SORIA**: Santa María de las Hoyas, monte “Sierra, Jabinada y otros”, 489656 E, 4621980 N, suelos calizos en sabinar de *Juniperus thurifera*, 1069 m, R. Belinchón & M. Prieto, 25/05/2006, M. Prieto 633. **ZARAGOZA**: Oseja, 607653 E, 4606638 N, sustrato yesíferos, suelo entre musgos, 837 m, I. Martínez & M. Prieto, 21/08/2007, M. Prieto 1090. **PORTUGAL**. Bragança, 29TPG799245, rocas básicas, anfíbolitas, 955 m, I. Martínez & M. Prieto, 06/09/2006, M. Prieto 838 (MA 16174).

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# MYCOTAXON

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## A new species of *Paradendryphiopsis* from Portugal

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**Abstract** — *Paradendryphiopsis pleiomorpha* sp. nov., found on the bark of an unidentified plant in Braganza, Portugal, is described and illustrated. It is distinguished by conidia that are catenulate, mostly 1–3-septate, usually ellipsoid or obclavate, navicular to oblong, smooth, with pale brown ends and brown at the middle, formed by blastic mode through the conidiogenous locus on unbranched, macronematous conidiophores and by a “thallic-arthric” *Bahusakala*-like synanamorph, which arises from the same conidiophores and vegetative hyphae. A key to *Paradendryphiopsis* species is presented.

**Key words** — systematics, anamorphic fungi

## Introduction

Ellis (1976) erected the genus *Paradendryphiopsis* for *P. cambrensis* M.B. Ellis (type species), found on dead wood of *Quercus* sp. in Wales. The author remarked that primary characteristics of the genus are monotretic conidiogenous cells and thin-walled, catenulate conidia. Hughes (1979) added a second species, *P. laxa* (H.J. Huds.) S. Hughes, and provided several illustrations and commentaries on

conidium ontogeny in *P. cambrensis*. Regarding *P. cambrensis*, Hughes (1979) wrote,

“Conidia are blastic rather than tretic as described, the deeply pigmented and conspicuous outer wall of the conidiogenous cell is constricted at its apex but entirely continuous with that of the conidium. Acropetal chains of two or three conidia are produced. When the conidium is mature the inner wall of the conidiogenous cell retreats somewhat from the apex and appears as a convex dome. Sometimes the base of the conidium may be temporarily attached, by means of a short denticle, to the retreated inner wall after the outer wall has already ruptured”.

Morgan-Jones et al. (1983) followed the same criteria when they described the third species, *Paradendryphiopsis anomala* Morgan-Jones et al., and treated the conidiogenous cells as monoblastic rather than tretic since continuity is clear between the wall of the conidiogenous cell and that of the conidium. During a November 2007 survey of microfungi in the Montesinho and Douro Natural Park (Portugal) as part of a mycological survey called “Flora Micológica Ibérica,” a conspicuous fungus from the genus *Paradendryphiopsis* was collected. The specimen showed differences from previously described taxa and is proposed as new to science.

## Materials and methods

Plant material was sampled during a mycological survey in the Montesinho Natural Park, Braganza, Portugal. Individual collections were placed in paper and plastic bags, taken to the laboratory, and treated according to Castañeda (2005) and Castañeda et al. (2010). Mounts were prepared in polyvinyl alcohol-glycerol (8 g in 100 ml of water + 5 ml of glycerol) and measurements made at 1000× magnification. Micrographs were obtained with a Zeiss Axio-Imager M1 light microscope.

## Taxonomy

***Paradendryphiopsis pleiomorpha*** R.F. Castañeda, Silvera, Gené & Guarro, **sp. nov.**

MYCOBANK MB 518830

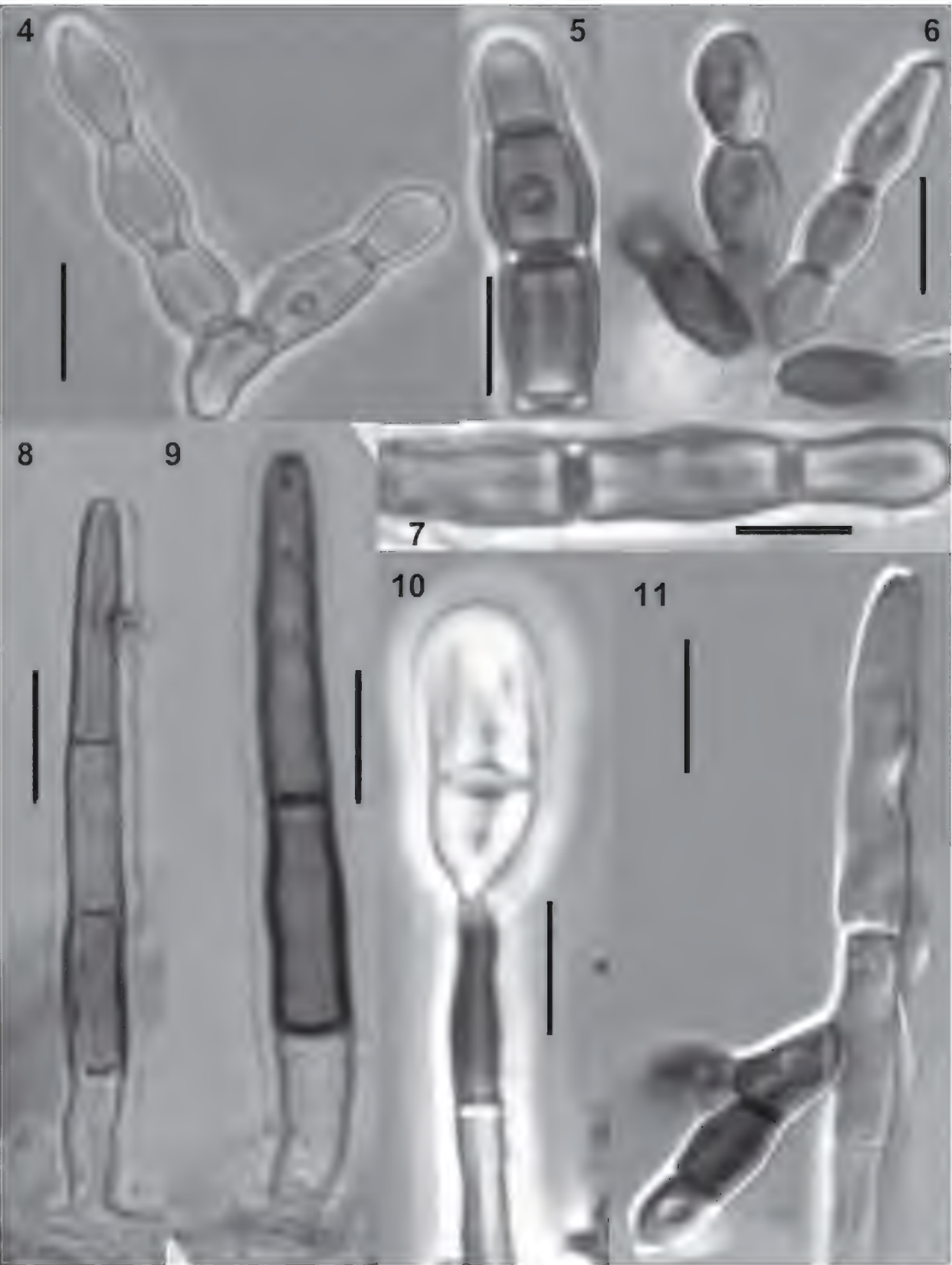
FIGS 1–14

*COLONIAE* in substrato naturali effusae, pilosae et funiculosae et interdum granulosaе, atrobrunneae. Mycelium partim superficial et partim in substrato immersum, ex hyphis septatis, ramosis, subhyalinis vel dilute brunneis, laevibus, 3–5 µm diam., compositum. *CONIDIOPHORA* mononematosa, macronematosa, simplicia, erecta, recta, cylindrica, 2–6-septata, laevia, irregulartim pigmentata, subhyalina vel dilute brunnea ad basim et brunnea vel dilute brunnea ad apicem, interdum fumoso-brunnea vel atrofumoso-brunnea, 40–150 × 4–6 µm. *CELLULAE CONIDIOGENAE* monoblasticae, terminales, determinatae, brunneae vel dilute brunneae, interdum fumoso-brunneae vel atrofumoso-brunneae, 25–40 × 4–5 µm. *CONIDIA* ellipsoidea, aliquot obclavata, ad usque oblonga, raro navicularia, blasto-catenulata, 1–3 septata, plerumque 2-septata, laevia, 17–30 × 6–9 µm, sicca, utrimque



FIG. 1-3. *Paradendryphiopsis pleiomorpha* photomicrographs from holotype (IMI 398786). Conidia and conidial chain. Scale bars = 10 µm.

*dilute brunnea et cellula centralis atrobrunnea, interdum irregulartim pigmentata cum unica cellula basalis vel apicalis dilute brunnea et cetero atrobrunnea vel atrofumoso-brunnea, praedita. SYNANAMORPHA ad genus Bahusakala similis, nonnumquam ipsis ex hyphis et conidiophoris exoriens cum conidiophoris micronematis, ramosis et irregulartim fasciculatis, ramoconidia et conidia “thallica-arthrica”, catenulata, per disarticulationem*



FIGS. 4–11. *Paradendryphiopsis pleiomorpha*, photomicrographs from holotype (IMI 398786). 4–7. Conidia of the *Bahusakala*-like synanamorph. 8–11. Conidiophores and conidiogenous cells, young attached conidium and *Bahusakala*-like synanamorph arising laterally from a conidiophore. Scale bars = 10 µm.

*ramorum producto, oblonga, doliiformia vel in forma plus minusve litterae Graecae upsilon, ex unicellularia, atrofumoso-brunnea vel atrobrunnea, laevia, sicca, 4–17 × 4–7 µm. Teleomorphosis ignota.*

TYPE: Portugal. Braganza, Montesinho Natural Park, on bark of an unidentified plant, 14.XI.2007. R.F. Castañeda, C. Silvera & J. Capilla (HOLOTYPE: IMI 398786; ISOTYPE: FMR 10132).

ETYMOLOGY: Greek, *pleio-*, meaning more than usual; *-morpha*, referring to existing forms of conidium ontogeny.

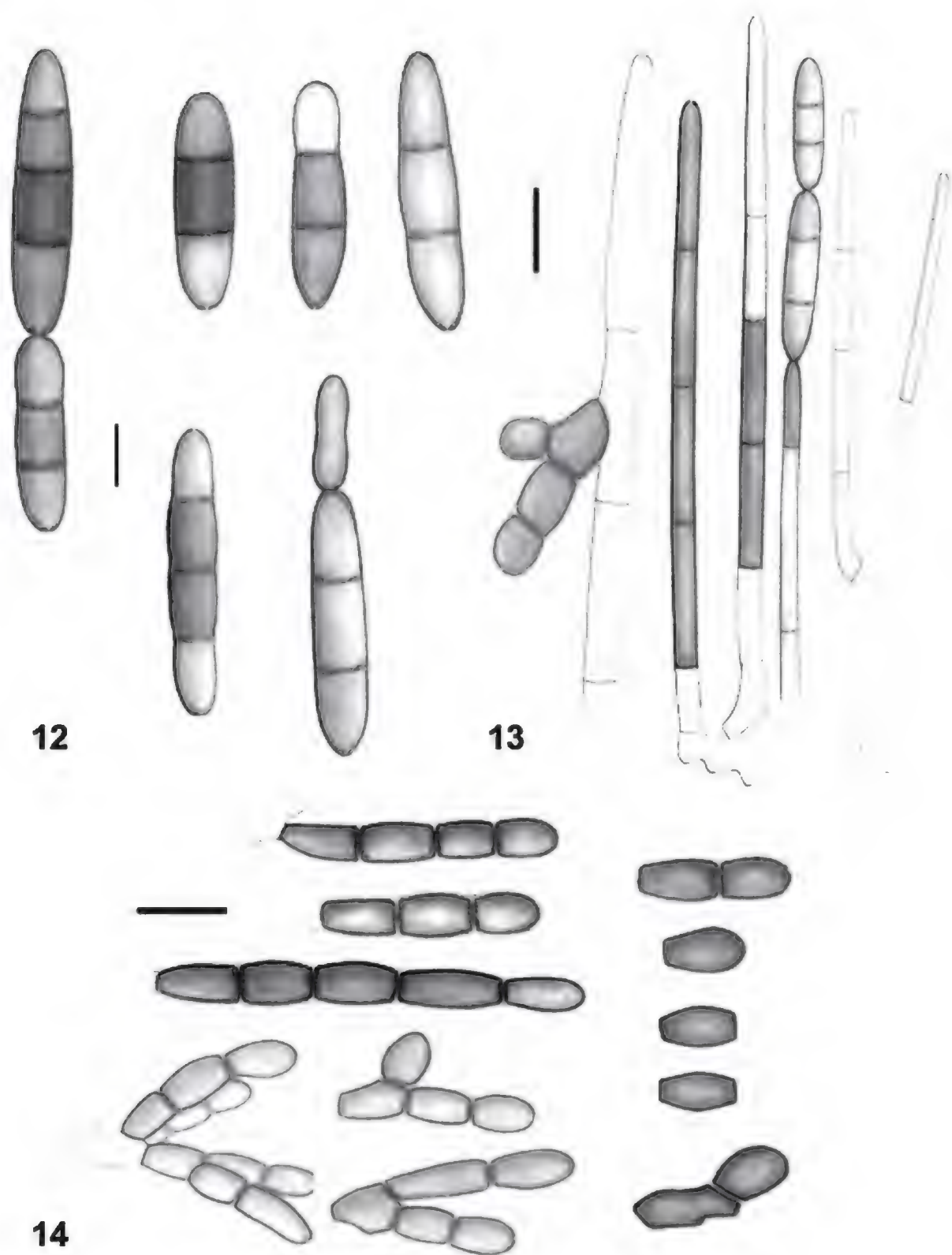
COLONIES on the natural substrate effuse, hairy and funiculose, sometimes granular, dark brown. Mycelium superficial and immersed; hyphae septate, branched, 3–5 µm diam., smooth-walled, subhyaline or pale brown. CONIDIOPHORES mononematous, macronematous, simple, erect, straight, cylindrical, 2–6-septate, smooth, subhyaline or pale brown at the base and brown or pale brown towards the apex, but sometimes irregularly pigmented grayish brown or dark grayish brown, 40–150 × 4–6 µm. CONIDIOGENOUS CELLS monoblastic, integrated, terminal, determinate, brown or pale brown, sometimes grayish brown to dark grayish brown, 25–40 × 4–5 µm. CONIDIA ellipsoid, somewhat obclavate, rarely navicular or oblong, blastocatenulate, 1–3-septate, mostly 2-septate, smooth-walled, 17–30 × 6–9 µm, dry, usually pale brown at the ends (sometimes only one end paler than the rest) and dark brown to dark grayish brown at the middle. SYNANAMORPH *Bahusakala*-like, arising from the same vegetative hyphae and conidiophores. Conidiophores micronematous, branched, irregularly fasciculate, dark brown to dark grayish brown. RAMOCONIDIA AND CONIDIA “thallic-arthric”, catenulate, oblong, doliiform, broadly Y-shaped, unicellular, dark gray-brown or dark brown, smooth, dry, 4–17 × 4–7 µm, forming by disarticulation of the conidiogenous branches. Teleomorph unknown.

*Paradendryphiopsis pleiomorpha* slightly resembles *P. cambrensis*, but that species has discrete conidiogenous cells and lacks a *Bahusakala*-like synanamorph. The pigment distribution in the conidiophores and conidia in that species is also quite distinct from *P. pleiomorpha* and can be easily differentiated (see key below).

#### Key to *Paradendryphiopsis* species

- 1 Conidiogenous cells discrete ..... 2  
Conidiogenous cells integrated ..... 3
- 2(1) Conidia ellipsoid, 3-septate, with end cells pale brown to subhyaline and intermediates ones brown, smooth, dry, blastocatenulate, 12–19 × 4–5 µm ..... *P. cambrensis*  
Conidia ellipsoid to clavate or turbinate, narrowed to truncate base, 2–3-septate, mid to dark brown, end cells pale, with dark brown bands at the septa, smooth, blastocatenulate dry, 16–30 × 8–12 µm ..... *P. laxa*





FIGS. 12–14. *Paradendryphiopsis pleiomorpha*, drawings from holotype (IMI 398786).  
12. Conidia. 13. Conidiophores, conidiogenous cells, conidia, and *Bahusakala*-like synanamorph arising from a conidiophore. 14. Conidiophores and conidia of the *Bahusakala*-like synanamorph.  
Scale bars = 10  $\mu$ m.

- 3(1) Conidia blastocatenulate, ellipsoid, somewhat obclavate, rare navicular or oblong, 1–3-septate, mostly 2-septate, smooth-walled, dry, pale brown at the ends, dark brown at the middle, sometimes irregularly pigmented, with basal or apical cell pale brown and dark brown to dark grayish-brown the rest,  $17\text{--}30 \times 6\text{--}9 \mu\text{m}$  ..... *P. pleiomorpha*  
 Conidia solitary, ellipsoid, smooth, 3–4-septate, brown, with the outer cells paler, usually slightly constricted at the end septa, dry, slightly truncated at the base,  $24\text{--}26 \times 11\text{--}13 \mu\text{m}$  ..... *P. anomala*

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We are deeply indebted to Prof. Lori M. Carris (Washington State University) and Dr. De-Wei Li (The Connecticut Agricultural Experiment Station) for kindly reviewing the manuscript. This study was supported by the Ministry of Science and Innovation of Spain, grant CGL 2008-04226/BOS. We thank the Cuban Ministry of Agriculture for facilities. The author RFCR thanks Drs Uwe Braun, Lori Carris, De-Wei Li, Felipe Wartchow, Antonio Hernández-Gutiérrez, Melissa Mardones, Cony Decock, Shaun Pennycook, Walter Gams, Roland Kirschner, Gabriela Heredia, Xiu Guo Zhang, D.J. Bhat, Gregorio Delgado, Eric H.C. McKenzie, and Pedro Crous for their generous and valued assistance with literature not otherwise available. We also acknowledge the facility provided by Dr. P.M. Kirk through the IndexFungorum website.

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**New records and checklist of corticioid *Basidiomycota* from Uruguay**SEBASTIÁN MARTÍNEZ<sup>1</sup> & KAREN K. NAKASONE<sup>2</sup><sup>1</sup>sebamart@fing.edu.uy*Laboratorio de Micología Facultad de Ingeniería/ Ciencias,  
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**Abstract** — Twenty-eight corticioid basidiomycete species are reported from Uruguay for the first time. An annotated checklist with 110 species of corticioid *Basidiomycota* recorded from Uruguay is presented based on these new records and an intensive literature search. These species are distributed in 49 genera and 10 orders. The order *Polyporales* is represented by the most species (40) and the genus *Phanerochaete* has the most species (11). *Hjortstamia fuscomarginata*, *Hyphoderma rimosum*, *Phlebia lividina*, and *P. subserialis* are recorded for the first time from South America. For the complete checklist see <http://www.mycotaxon.com/resources/weblists.html>.

**Key words** — biodiversity, *Homobasidiomycetes*, taxonomy, wood-rot

**Introduction**

Uruguay is located in southeast South America between 30° and 35°S and 53.5° and 58.5°W, covering around 178,000 km<sup>2</sup>. The mean temperature is 17.5°C varying from 16°C in the southeastern Atlantic coast to 19°C in the northwest. The mean annual precipitation is 1300 mm, ranging from 1100 mm in southern Uruguay to 1600 mm in the north (Dirección Nacional de Meteorología 2009). The climate in Uruguay is rainy, without a dry season, but with a wide annual variation in precipitation. The Uruguayan climate is temperate and wet (type “C”) with precipitation throughout the year (type “f”); in the hottest month the temperature is over 22°C (type “a”) (Dirección Nacional de Meteorología 2009). These characteristics correspond with the Cfa climate type of the Köppen-Geiger classification (Peel et al. 2007).

In Uruguay, 7% is forested and 80% is grasslands (Carrere 2001). About 750000 ha are covered by native forests and an additional 670,000 ha consist

of nonnative forests of mostly *Eucalyptus* and *Pinus* species for the pulp and sawmill industries (Anon. 2005). The native vascular flora of Uruguay consists of approximately 2500 to 2750 species (Marchesi 2005, Alonso-Paz & Bassagoda 2002) including 302 indigenous tree and shrub species (Brussa & Grela 2007). According to Alonso-Paz & Bassagoda (2002), the Uruguayan vascular flora is composed of 150 families and 859 genera, which is high if measured by unit area. The families with the highest number of species are *Asteraceae*, *Poaceae*, *Fabaceae*, *Cyperaceae*, and *Euphorbiaceae* (Marchesi 2005, Alonso-Paz & Bassagoda 2002). This diversity of woody native and introduced plant species suggest a corresponding high level of fungal diversity.

The corticioid Basidiomycetes of Uruguay are poorly known. Felippone (1928) was the first to record corticioid species from Uruguay. He recorded four species of *Thelephora* and eight in *Stereum*. Herter (1933) reported six species of *Thelephoraceae*, including one species of *Hymenochaete* and two species in *Irpex* and *Merulius*. Koch et al. (1981) recorded eight species in the genera *Corticium*, *Stereum* and *Thelephora*, as related to plant pathology. In a series of papers, Gazzano (1987, 1988, 1990, 1992, 1994, 1996, 1998, 2000, 2001, 2002, 2007) reported on various polyporoid and corticioid species from Uruguay, including many new records. In total, there are about 70 species of corticioid fungi reported from various sources. Recent collections from throughout Uruguay on native and exotic trees yielded new records of corticioid basidiomycetes. In this study, we report an additional 28 new records of corticioid species. The aim of the present work is to establish a baseline of knowledge of the diversity of corticioid basidiomycetes in Uruguay by providing a checklist of the recorded species.

### Materials and methods

The checklist is based on data obtained from an intensive search of literature records of corticioid fungi from Uruguay. Genera and species are listed alphabetically within each accepted order according with the proposed nomenclature of Hibbett et al. (2007) and Larsson (2007). Data on substrate and nutritional strategies are provided for each species. The new species records in this study were collected in native and nonnative, planted forests, urban areas, or retrieved from the herbarium of the Facultad de Ciencias, Montevideo, Uruguay (MVHC). Microscopic examinations were made from freehand sections mounted in 5% aqueous KOH and 1% aqueous phloxine solutions, 5% cotton blue in 25% lactophenol, and Melzer's reagent (Kirk et al. 2008). Specimens were deposited at MVHC. Author abbreviations follow Kirk & Ansell (1992). Cortbase version 2.1 (Parmasto et al. 2004) and Index Fungorum ([www.indexfungorum.org](http://www.indexfungorum.org)) were consulted for current names of species and synonyms.



## Results

The corticioid basidiomycetes of Uruguay consist of 110 recorded species, including the present additions. Ninety-nine species are here taxonomically or nomenclaturally accepted and eleven are listed as doubtful. These are distributed in 10 orders according with the modern classification based on molecular studies (Hibbett et al. 2007, Larsson 2007). Among them, only three species belonging to the *Boletales* are brown-rot decay fungi. The orders with the highest number of species present in Uruguay are *Polyporales* (40 species), *Hymenochaetales* (25 species), and *Russulales* (16 species). The remaining seven orders are represented by five or fewer species. The genera with the highest number of recorded species are *Phanerochaete* (11 species), *Phlebia* (8 species) and *Hyphodontia* (7 species) from a total of 49 genera represented in the Uruguayan checklist. *Hjortstamia fuscomarginata* (Burt) Hjortstam & Ryvarde, *Hyphoderma rimosum* Burds. & Nakasone, *Phlebia lividina* Hjortstam and *P. subserialis* (Bourdot & Galzin) Donk are recorded for the first time from South America. For the complete checklist see <http://www.mycotaxon.com/resources/weblists.html>.

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## BOOK REVIEWS AND NOTICES

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## INTRODUCTION

Monographic systematic treatments of diverse fungal groups are the focus of this installment of BOOK REVIEWS AND NOTICES. The first four reviews cover four volumes in the series STUDIES IN MYCOLOGY that focus on different groups of *Ascomycota*. The other five publications have *Agaricales* and *Russulales* as the subject. A worldwide overview of the species and genera in the *Xerula/Oudemansiella* complex, *Lactarius* in Africa, and European representatives of the genera *Hygrocybe*, *Conocybe* and *Pholiotina*, and *Agaricus* are reviewed. These books, although with a regional approach, have a much wider usability than only for the region for which they were researched and written. Most of these books are lavishly illustrated with colour pictures, thanks to today's digital cameras and the modern printing techniques. The Internet with its resources and digitalized texts means that mycology is no longer only a privilege for those with access to well-stocked libraries. The two books in the series FUNGI EUROPAEI that are reviewed here are examples of this democratization process, as both authors are not mycologists by profession: the author of the *Agaricus* book is a practicing veterinarian. It seems fitting that he explicitly acknowledges the on-line sources for old(er) mycological literature.

This contribution concludes with a list of newly published books to be included in upcoming BOOK REVIEWS AND NOTICES.

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<sup>1</sup> Books for consideration for coverage in this column should be mailed to the Book Review Editor at the address above. All unsigned entries are by the Book Review Editor.



## ASCOMYCETES

**A phylogenetic re-evaluation of *Dothideomycetes*.** By C.L. Schoch, J.W. Spatafora, H.T. Lumbsch, S.M. Huhndorf, K.D. Hyde, J.Z. Groenewald & P.W. Crous. 2009. STUDIES IN MYCOLOGY no. 64. CBS Fungal Diversity Centre, PO Box 85167, 3508 AD Utrecht, The Netherlands. <info@cbs.knaw.nl>. Pp. vi + 220, illustr. ISBN 978-90-70351-78-6. Price: 65 €.

The taxonomy of the dothideomycetous fungi, i.e. most of those with bitunicate asci, has been in a state of continuous flux for over a century, with vastly different systems being proposed by some and then overturned by others. Part of this difficulty has been a consequence of how particular characters should be interpreted and weighted, but even more of a problem has been the vastness of the group, which makes it difficult for a single mycologist to appreciate the breadth and complexity of the included fungi – both morphologically and biologically. The most significant morphologically based works on the group in the last quarter of the 20th century have been the generic keys and compilation of synonyms by von Arx & Müller (1975), the critical studies on the types of family names by Eriksson (1981), and the illustrated overview of families and genera by Barr (1987). The present issue is fittingly dedicated to the three of those now deceased. However, all these authors adopted different systems of orders and families, and development of a robust classification has only become feasible with the advent of molecular phylogenetic methods. In such a complex group, inadequate sampling, even at the ordinal level, has meant that molecular phylogenies have also been in flux. Indeed, it is only in the last few five years that a more stable backbone has started to emerge as the representation of families and genera has improved. The present volume evidences the enormous and exciting progress that has been made, but simultaneously reveals areas of continuing uncertainty and instability where yet more work is required.

The scene is set by a five-gene phylogeny derived from 356 isolates representing 41 families (of which six are newly described elsewhere in the volume) and all currently accepted orders. Prepared by Schoch and 53 co-authors, this also includes an analysis of the biology of the taxa, leading to the somewhat contentious view that there have been numerous transitions from saprobic to plant-associated and lichenized life-styles. However, a genome-level comparison revealed a high level of unique protein coding genes in the class compared with other fungi, supporting the recognition of *Dothideomycetes* as a distinct class. The major part of the volume, however, is devoted to more detailed studies of particular orders, families, or representatives with different biologies or ecologies.

The monophyly and family structure in *Capnodiales* is addressed by Crous et al., where the main surprise is the placement of *Piedraiaceae* inside *Teratosphaeriaceae*; the new family *Dissoconiaceae* is also proposed. The families and genera of the former *Hysteriales* are revisited in a multi-gene phylogeny by Boehm et al.; *Hysteriales* is supported as sister to *Pleosporales*, while *Mytilinidiales* (including *Gloniaceae*) is sister to both. Here a particularly surprising find was that the asexual *Cenococcum geophilum* falls in *Gloniaceae* – a result that may merit more critical scrutiny. In the case of *Pleosporales*, Zhang et al. compared five loci in representatives of 59 genera and 15 families; two new families (*Amniculicolaceae*, *Lentitheciaceae*) are introduced, *Pleomassariaceae* is included in *Melanommataceae*, and the familial positions of several genera are clarified. Mugambi & Huhndorf revisit the *Melanommataceae* and *Lophiostomataceae* issue; while both families and *Hypostromataceae* were recovered, *Melanommataceae* and, in particular, *Melanomma* remain polyphyletic — however, *Bertiella* and *Herpotrichia* did belong to that family, and an atypical new genus *Misturatosphaeria* is described.

The problem of unnamed lineages recovered from rock is explored further by Ruibal et al., who again emphasize the phylogenetically diverse positions of these superficially rather similar fungi; they include representatives of four dothideomycete orders but one lineage appears closer to *Arthoniomycetes* — many more of these rock-inhabiting fungi clearly remain to be found, and at least the main lineages will eventually have to be named as new genera, even in the absence of either a sexual or an asexual stage, if no already named fungi sequenced continue to prove to be distinct. Nelsen et al. treat the lichenized representatives of *Dothideomycota* based on nuLSU and mtSSU sequence data; *Arthoniomycetes* and *Dothideomycetes* are supported as separate classes; the study shows that in several cases generic concepts require revision, while *Mycomicrothelia* (a genus which includes both lichenized and non-lichenized species) is found to be sister to *Trypetheliaceae* rather than a member of *Arthopyreniaceae*. Shearer et al. studied 169 freshwater isolates, of which 84 belonged in *Dothideomycetes*; within the four clades including only freshwater species — *Jahnulales* the largest (the others being *Lingoldiomycetaceae*, *Amniculicolaceae*, and *Tingoldiogo* + allies) — the aquatic habit is regarded as secondary, all having terrestrial ancestors. Suetrong et al. reached similar conclusions for marine *Dothideomycetes*, which were found to be dispersed through 12 families in six orders in a four-gene phylogeny; most occur on intertidal plants and are tropical, with novel taxa continuing to be recognized, which include two new families (*Aigialaceae*, *Morosphaeriaceae*) and three new genera introduced here. Finally, Tanaka et al. propose the new family *Teratosphaeriaceae* for five new genera of *Massarina*-like bambusicolous fungi

with *Tetraploa* and *Tetraploa*-like anamorphs or which only produce conidia; here the beautiful *Quadricrura* has species with 1–2 long apical and 4–5 short more basal setae.

The whole issue is illustrated by stunning top-quality and artistically composed colour photomicrographs, and also colour-coded phylograms, which greatly facilitates their interpretation. There is no doubt that this will be regarded as a classic work on the class (!), but I was disappointed that only one chapter (Boehm et al.'s on the hysterioid groups) included any keys. Keys to families, and at least the genera and species treated in detail, would have made the work much more accessible to those wishing to use this volume in making identifications using microscopic characters. Mycologists with access to superbly equipped and resourced molecular laboratories, supported by skilled technicians, should not forget that they represent a privileged section of the potential user-community of systematic works.

Arx JA von, Müller E. 1975. A re-evaluation of the bitunicate ascomycetes with keys to families and genera. *Studies in Mycology* 9: 1–159.

Barr ME. 1987. *Prodromus to Class Loculoascomycetes*. Amherst, MA: ME Barr.

Eriksson OE. 1981. The families of bitunicate ascomycetes. *Opera Botanica* 60: 1–220.

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**Highlights of the *Didymellaceae*: a polyphasic approach to characterise *Phoma* and related pleosporalean genera.** By M. Aveskamp, H. de Gruyter, J. Woudenberg, G. Verkley & P.W. Crous. 2010. *STUDIES IN MYCOLOGY* no. 65. CBS Fungal Diversity Centre, PO Box 85167, 3508 AD Utrecht, The Netherlands. <info@cbs.knaw.nl>. Pp. iv + 64, illustr. ISBN 978-90-70351-79-3. Price: 40 €.

With over 3200 species names in Index Fungorum/MycoBank, *Phoma* is surely one of the largest morasses requiring resolution amongst the microfungi. This slim volume does not have all the answers, but makes important inroads into identifying the directions of future work by re-assessing the nine-section morphology based system of Boerema et al. (2004; see MYCOTAXON 90: 487–492, 2004); the sections in that system were separated by differences in pycnidial wall anatomy, the occurrence of setae, conidium size, and the presence of chlamydospores. In this issue of *STUDIES*, a commendable 324 strains are compared by molecular phylogenetic methods, representing 206 taxa of which 159 are *Phoma*-like. Eighteen clades are recognized which, perhaps not surprisingly, do not correlate with the earlier sectional system. Just four of those clades — ones that could be separated morphologically



— are named here: *Didymella* (incl. *Phoma herbarum*, the type species of *Phoma*), *Boeremia* gen. nov. (for *P. exigua* and allied species), *Peyronella* (with teleomorphs formerly in *Didymella* and *Mycosphaerella* — controversially combined under the anamorphic name), and *Stagonosporopsis* (for the former *Phoma* sect. *Heterospora*). In addition, the anamorphs of *Leptosphaerulina* and *Macroventuria* came together in another of the 18 clades. No links with any true *Mycosphaerellaceae*, or indeed any group in *Capnodiales*, were upheld. Sixty-one new combinations are made, and eight new species and two new varieties are described in addition to the new genus.

In view of the limited representation of the treated species, almost all of which are from plants and known in culture, it will be interesting to see whether there is any change in the support for the clades found here when specimens from other host plants, and such disparate hosts as lichens, can be incorporated into the analysis. In the meantime, those working with the untreated species will have to be content to continue to use the current morphology based circumscription of *Phoma*, but in doing so should also appreciate that they are being pragmatic and using the name ad interim in a polyphyletic sense.

Boerema GH, de Gruyter J, Noordeloos ME, Hames MEC. 2004. *Phoma* Identification Manual: differentiation of specific and infra-specific taxa in culture. CABI Publishing, Wallingford.

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**Systematics of *Calonectria*: a genus of root, shoot and foliar pathogens.** By L. Lorenzo, P.W. Crous, B.D. Wingfield & M.J. Wingfield, 2010. STUDIES IN MYCOLOGY 66. CBS Fungal Biodiversity Centre, Uppsalalaan 8, 3584 CT, Utrecht, the Netherlands. <info@cbs.knaw.nl>. Pp. iv + 71, illustr. ISBN 978-90-70351-81-6. Price: 40 €.

The approach taken in this taxonomic revision may be controversial for nomenclatural pedants, but it is pragmatic in a time when changes in the International Code of Botanical Nomenclature relating to the separate naming of states of pleomorphic fungi may be imminent. They treat the generic name *Cylindrocladium*, which is typified by a conidial state fungus, as a regular synonym of *Calonectria*, which is based on a sexual state; i.e. they apply the one-name-for-one-fungus approach, which can only be welcomed by those working with these fungi. The proclamation that “new species should be described in *Calonectria* irrespective of whether the teleomorph is known or not” (p. 3) is pragmatic in this case, where there is a complete congruence between the circumscriptions of the two genera.

The issue comprises three contributions. First is a discussion of species concepts and the nomenclatural approaches adopted, also emphasizing the importance of the genus as plant pathogens. Second is what might be seen as an exemplar study of the plant pathogenic *Calonectria pauciramosa* s. lat. in which a multigene phylogeny and mating tests demonstrate the occurrence of three previously unrecognized cryptic species, which are here described as new. But it is the third that will be of especial interest to those concerned with identification of fungi in the genus – a multigene phylogeny and synopsis that accepts a total of 68 species, of which seven are new to science, and 18 new combinations (all with basionyms in *Cylindrocladium*). Diagnostic characters of the conidial states are illustrated by photomicrographs, and most pleasing are the synoptic and dichotomous keys to the 68 species now accepted under *Calonectria* (i.e. including *Cylindrocladium*). While this is no monograph with detailed descriptions and information on hosts and distributions (as the authors recognize on p. 10), the issue will facilitate the accurate identification of these fungi by plant pathologists and citizen scientists. All concerned with these fungi will need to have this to hand, at least until a full monographic treatment becomes available.

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**Species and ecological diversity within the *Cladosporium cladosporioides* complex (*Davidiellaceae*, *Capnodiales*).** By K. Bensch, J.Z. Groenewald, J. Dijksterhuis, M. Starink-Willemse, B. Andersen, B.A. Summerell, H-D Shin, F.M. Dugan, H-J Schroers, U. Braun & P.W. Crous, 2010. *STUDIES IN MYCOLOGY* 67. CBS Fungal Biodiversity Centre, Uppsalalaan 8, 3584 CT, Utrecht, the Netherlands. <info@cbs.knaw.nl>. Pp. iv + 96, illustr. ISBN 978-90-70351-83-0. Price: 50 €.

Our understanding of the taxonomy of the remarkably successful fungi referred to *Cladosporium* has advanced dramatically over the last few years as more and yet more isolates have been studied by molecular phylogenetic methods – as witnessed by a previous number of *STUDIES* devoted to the genus, its dismemberment, and also revisions of species concepts in *C. herbarum* and *C. sphaerospermum* (Crous et al. 2007; see *MYCOTAXON* 107: 507–509, 2009). This new number of the *STUDIES* might be viewed as a continuation or supplement to that of 2007 in addressing *C. cladosporioides* — a name widely used for saprobic fungi of the genus occurring on decaying or diseased plant parts and well-known as a spoilage and indoor mould growing on materials such as damp plasterwork. Now, over 200 isolates of the complex have been



analyzed by a multigene approach — resulting in an explosive expansion of the group. While the precise application of the name *C. cladosporioides* is fixed here by neo- and epitypification, a staggering 22 species are described as new to science. Although recognized as a result of molecular studies, diagnostic micromorphological features were found: differences in the shape, width, length, septation, and surface ornamentation of the conidia and conidiophores; the length and branching patterns of conidial chains; and hyphal shape, width, and arrangement. The surface features of the conidia were examined using Cryo-SEM and the conidia were found to have a characteristic reticular or embossed striped ornamentation. All these features are seen in the superb photomicrographs provided, which leave no doubt that there are non-molecular characters of value, even though very careful comparisons will often be required.

I was very pleased to see that a dichotomous key had been provided, and that the couplet characters were almost all morphological or micromorphological. However, variability has necessitated that several species were keyed out more than once and, somewhat frustratingly, no micromorphological features were found to distinguish some of the novel phylogenetic species closest to *C. cladosporioides* s. str., so that after the couplet leading to that name placed in parenthesis is “(including morphologically indistinguishable but phylogenetically distinct lineages).” The implication of this is that, without molecular sequence data, it is no longer possible to recognize *C. cladosporioides* s. str., which means that morphological identifications will have to have appended “complex” or “s. lat.” A further complication is that instances were found where several isolates from a single location and precise substratum (e.g., an individual plant) yielded more than one widely separated species of the complex. The phenomenon of co-occurrence of different species of *Mycosphaerella* and *Teratosphaeria* in the same leaf lesions has previously been documented, so this result is perhaps not surprising, but it does mean that enormous care is needed in isolating these fungi from natural habitats to be confident that representative lineages have been obtained. This revision has consequently elegantly clarified the species concepts in this group of economically important fungi, but simultaneously made it more difficult for some of the members now known to be in the complex to be identified in the absence of molecular data.

Crous PW, Braun U, Schubert K & Groenewald JZ (2007) The genus *Cladosporium* and similar dematiaceous hyphomycetes. *Studies in Mycology* 58: 1–253.

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## AGARICALES AND RUSSULALES

**The *Xerula/Oudemansiella* complex (Agaricales).** By R.H. Petersen & K.W. Hughes. 2010. BEIHEFTE NOVA HEDWIGIA 137. J. Cramer in der Gebr. Borntraeger Verlagsbuchhandlung, Johannesstraße 3A, 70176 Stuttgart, Germany. <mail@schweizerbart.de>. Pp. 625, plates 31, figs 576. No ISBN number. Price 179.00 €

The complex of the agaricoid genera *Xerula* and *Oudemansiella* (*Physalacriaceae*, *Agaricales*) is unraveled in great detail in this taxonomic treatment by Ron Petersen and Karen Hughes. The 625-page thick book reveals a much greater complexity than ever imagined. The complex is morphologically studied, and ITS and LSU phylogenies are constructed.

Let us first look at the contents of the book. After a general introduction with a history of the genera/genus and its classifications, material and methods for the research are given, followed by a chapter on the DNA-based phylogenies. 330 pages are devoted to genus and species descriptions, keys to the species, line drawings, and photographs. The next 200 or so pages contain the type studies, and finally a list of new taxa and new combinations, indices, and literature references fill the rest of the pages.

A big problem faced by the authors was how to name the supraspecific taxa, and whether to recognize one genus or name the separate clades. The choice was made to split the group and to recognize seven genera, four of them newly described here, some of them distinctly not monophyletic, but morphologically distinct and homogeneous. The two genera with non-rooting fruitbodies that grow directly on wood are *Oudemansiella*, restricted to tropical species without a persistent annulus, and *Mucidula* as the temperate counterpart with a persistent annulus. Although the two look very much alike, they are not sister groups. The other five genera all have a 'rooting' stipe connected to subterraneous wood or tree roots. The old *Xerula* is redistributed into *Xerula* (in the strict sense) for species with thick-walled setae on the pileus; *Paraxerula* harbours species with thin-walled setae on the pileus; *Hymenopellis*, with the highest number of species, is characterized by a moist to glutinous pileus; *Protoxerula* species, also with a sticky pileus, occur in Australia and have green colours; species with spiny spores are accommodated in the genus *Dactylosporina*; and *Ponticulomyces* (which did not make it into the general key) is an Asian clade of two species with characters in between *Hymenopellis* and *Oudemansiella*. *Hymenopellis* is not a monophyletic unit, and several other genera are nested within it; which genera depend on which gene region the phylogeny is based. The position of *Mucidula* in the middle of *Hymenopellis* was not expected. It is surprising that the authors have not tried to show more support for these decisions by either analyzing the data with topological constraints (such as a

monophyletic *Hymenopellis*) or adding data from protein coding genes. Another solution might be to recognize three genera — *Xerula* s. str. and *Paraxerula* as defined above plus *Oudemansiella* containing all other taxa, including the secotioid genus *Cribbea*. All three form well supported monophyletic clades in the ITS and the LSU phylogenies. Personally, I find the recognition of non-monophyletic genera very problematic, and this is my main critique on this book.

Besides the four new genera, four new species are described, one from Guyana, one from the USA, a third from India, and the fourth from eastern Russia.

The value of this monograph lies in the very thorough descriptions, not only of all accepted taxa, but also and especially of all the type specimens that could be studied. It is also extremely pleasant to have all this information in one place, and not scattered over various publications in a diverse set of journals. However, the information on the type collections should be searchable on the web, ideally linked to nomenclatural data, such as in Index Fungorum or Mycobank. On the negative side is of course the cost of this book, a high price that will certainly deter people in less developed countries from purchasing. This is very infelicitous, as the highest diversity of these taxa is in Asia.

The quality of the photos is variable, and some have been reproduced in a strange way. Unfortunately, but understandably, not all taxa are depicted with a colour plate.

With a book of this size it is inevitable that details have been overlooked; one Latin description never got beyond the first phase of some jotted down characteristics, the epithet 'kuehneri' is consistently misspelled as 'kuehnerii', and diacritical signs in non-English article titles and publications are not or wrongly applied.

This book should nonetheless find a wide audience due to its thorough descriptions and worldwide coverage.

***Agaricus* L. *Allopsalliota* Nauta & Bas. FUNGI EUROPAEI 1. 2<sup>nd</sup> Ed. By L. A. Parra Sánchez. 2008. Edizioni Candusso, Via Ottone Primo 90, 17021 Alassio SV, Italy. <maxcandusso@libero.it>. Pp. 824, Plates 396 + 42, figs 114. ISBN 88-901057-7-1. Price 75.00 €.**

This new book in the series FUNGI EUROPAEI replaces the 1984 and first volume on the genus *Agaricus* in the series on European fungi. This volume consists of a thorough and well-illustrated introduction to the genus, keys to the subsections, and extensive descriptions of and notes on the 35 species and varieties in sections *Agaricus*, *Bivelares*, *Chitonioides*, *Sanguinolenti*, and *Spissicaules*. The other two sections, viz. *Minores* (with subsections *Minores* and *Arvenses*) and

*Xanthodermatei*, the subgenus *Lanagaricus*, and the genus *Allopsalliota* will be covered in Part 2 that was scheduled to appear in 2009/2010, but one which we are still eagerly awaiting.

The book was written in Spanish with an English translation, and an Italian translation of the keys is also provided, which partly explains the volume of it. It is lavishly illustrated, with line drawings of the microscopical characters, numerous photos — always several per species showing the variability and changes the fruitbodies go through during maturation, and photos of micromorphological characters. Important characters are often separately illustrated, and photos of spot tests made with various chemicals are given as well.

The introduction alone is reason to buy this book: all you ever wanted to know about *Agaricus*, and much more, is covered. The overview of the characters that are used in *Agaricus* classifications and species recognition is excellent, with many colour photos to illustrate them.

Original diagnoses and plates are reproduced, either in black and white in the text or at the end as colour plates. This is a very valuable asset of this whole series.

Tables compare spore sizes by different authors for the taxa or give comparisons of closely related species.

This book is extremely well researched and executed. Although the European taxa are the focus of the book, its usage exceeds this area, for several reasons. First of all, it provides a clear concept of the European species, and secondly, mushroom species do not read maps and are not constrained by political boundaries. It is also very fortunate that the author has teamed up with those *Agaricus* researchers who apply molecular-phylogenetic methods to the genus for species recognition and circumscription.

A small comment I have is that it would have helped the user to have headers with the species names on top of the pages.

The happy spores on page 367 reflect my feelings when browsing through this book. The only thing missing is the mushroom smells...

Cappelli, A., 1984. *Agaricus* L. : Fr. (*Psalliota* Fr.) *Fungi Europaei* 1. Libreria editrice Biella Giovanna, Saronno.

***Conocybe* Fayod. *Pholiotina* Fayod. FUNGI EUROPAEI 11.** By A. Hausknecht. 2009. Edizioni Candusso, Via Ottone Primo 90, 17021 Alassio SV, Italy. <maxcandusso@libero.it>. Pp. 968, plates 46 + 403, figs 150, maps 154. ISBN 88-901057-8-X. Price 79.00 €.

Another thorough and excellent monograph in the FUNGI EUROPAEI series, volume 11 harbours all European taxa of *Conocybe* and *Pholiotina*. After the classic but of course heavily outdated 1935 book on the genus *Galera* by Kühner

and the much more recent work on the Dutch species by Arnolds (2005), this will be the book for the future on all aspects of these two genera. In almost 1000 pages, the 101 *Conocybe*, and 26 *Pholiotina* species are described, compared with each other, and illustrated with colour photos, watercolours, and black-and-white microdrawings. Little maps show in which European countries the species were found. As in the other volumes in the series, the original descriptions are reproduced and type studies are provided. The book starts out with an extensive introduction to the two genera, covering the history, classifications, and an overview of the main characters. This introductory text is in three languages: English, Italian, and German. The keys and descriptions of the supra-specific taxa are also trilingual, species descriptions are in English, and comments are in English and German. The list of examined collections also notes whether that particular collection is depicted in the literature, a feature I have not seen elsewhere.

Thickness and colour of the spore wall turn out to be very important in the identification, and it is a pity that those characters are not depicted. The line drawings fall short here (the ones in Arnold's work are of better quality), and colour photos would have been more helpful.

The author also contributed to the sections on the two genera in FUNGA NORDICA (2008), but the present work covers a much wider area and more species. With the relatively low cost of this book, it should find its way to many mycologists' bookshelves.

Arnolds E. 2005. *Conocybe. Pholiotina*. In Noordeloos ME, Kuyper ThW, Vellinga EC (eds). Flora agaricina neerlandica 6: 120–203. Taylor & Francis, Boca Raton, etc.

Hausknecht A, Vesterholt J. 2008. *Conocybe; Pholiotina*. In Knudsen H, Vesterholt J (eds). Funga Nordica: 626–645; 651–657. Nordsvamp, Copenhagen.

Kühner R. 1935. Le genre *Galera* (Fries) Quélet. Lechevalier, Paris.

**The genus *Hygrocybe*.** 2<sup>nd</sup> revised edition. FUNGI OF NORTHERN EUROPE Vol. 1. By D. Boertmann, 2010. Danish Mycological Society, Søvnøget 9, 3100 Hornbæk, Denmark. <svampetryk@webspeed.dk>. Pp 200, colour plates, line drawings, distribution maps. ISBN 978-87-983581-7-6. Price DKK 280

The second edition of this handsome book, in which the *Hygrocybe* species from northern Europe are depicted and described, shows some significant changes in comparison with the first (Boertmann 1995), now out of print: it is in hardcover, and three additional taxa are treated, many new colour plates of these bright and beautiful fungi are added showing more than ever the extreme colour variability, and the introduction and references are updated. Not yet updated are the genus names that might have to be adopted because of the progress in phylogenetic studies based on DNA comparisons. *Hygrocybe* in the sense presented here is not monophyletic. Some species are better placed in



*Omphalina/Arrhenia*, outside the *Hygrophoraceae*, *Cuphophyllus* (also known as *Camarophyllus*) and *Gliophorus* are well characterized genera within the *Hygrophoraceae*, but as there is not yet a thorough molecular-phylogenetic analysis of the family as a whole, these decisions have been postponed. Three new combinations that were invalidly introduced in FUNGA NORDICA (Boertmann 2008) are here validated.

The photos are just plain beautiful and in themselves a reason to buy this book. This book is particularly valuable for all who are trying to survey, manage and conserve the vulnerable unfertilized grasslands in (northern) Europe, and the author mentions, with pride, a British court case in which the presence of wax caps stood in the way of building developments. Of course, this book can be used in a much larger area than just northern Europe; it gives well-illustrated descriptions of the European species whose names are widely applied elsewhere.

Boertmann D. The genus *Hygrocybe*. Fungi of northern Europe vol. 1. The Danish Mycological Society.

Boertmann D. 2008. *Hygrocybe* (Fr.) P. Kumm. In Knudsen H, Vesterholt J (eds). *Funga Nordica*: 194–212. Nordsvamp, Copenhagen.

**Fungus flora of tropical Africa. Volume 2. Monograph of *Lactarius* in tropical Africa.** By A. Verbeken & R. Walley. 2010. National Botanic Garden of Belgium, Nieuwelaan 38, 1860 Meise, Belgium, <sales@br.fgov.be>. Pp. 151, plates 54. ISBN 978-90-726-1981-5. Price 50.00 €.

Isolated early from the other continents and bounded to the north by the Sahara Desert, the African tropical forests possess an ectomycorrhizal mycota that is largely — perhaps completely — endemic (Verbeken & Buyck 2002). For over three-quarters of a century, the National Botanical Garden of Belgium has fostered the scientific knowledge of ectomycorrhizal and other macromycetes in central Africa through collecting expeditions and the publication series FLORE ICONOGRAPHIQUE DES CHAMPIGNONS DU CONGO (18 volumes; 1935-1972) and FLORE ILLUSTRÉE DES CHAMPIGNONS D'AFRIQUE CENTRALE (17 volumes; 1972-1997). A new series, the FUNGUS FLORA OF TROPICAL AFRICA (2007-present), represents a continuation of the two previous series. In the second volume of the FUNGUS FLORA OF TROPICAL AFRICA, Professor Annemieke Verbeken of Ghent University (Belgium) and the late Ruben Walley (1966-2008) present a monographic study of the genus *Lactarius* (*Basidiomycota*, *Russulales*) in tropical Africa.

Outside of Heim's (1938, 1955a, b) studies in Madagascar, Congo, and Western Africa, studies of *Lactarius* in tropical Africa were restricted to scattered species

descriptions until the early 1990s. In 1993, the authors began focused studies on *Lactarius* in this region, and the present volume compiles a substantial amount of knowledge about the topic. Verbeken and Walley present descriptions of 96 species and 2 accepted varieties within 17 subgeneric sections, with taxonomic keys to tropical African species provided for each section. A detailed, 20-page section describing taxonomically valuable characters is richly illustrated with line drawings of micromorphological features. Species descriptions are detailed and accompanied by exceptional line drawings. Eighty of the species are represented within the 54 full-page color plates by photographs, watercolors by M. Goosens-Fontana (whose striking watercolors appear in the previous two publication series), or both. The color photographs (mostly by the authors, B. Buyck, or A. De Kesel) are impressively large (most are half-page scale – significantly larger than those in most field guides, not to mention other monographs) and nearly all of them are of excellent quality; both characteristics combine to make the plates a valuable source of visual information. References, a taxonomic index, and French translations of the taxonomic keys are provided. At a list price of 50 € (\$68 US), this volume is quite reasonably priced given the number of photographs, and demonstrates that it is indeed possible to publish richly illustrated yet affordable taxonomic texts.

Though recent molecular systematic studies have established the non-monophyly of *Lactarius*, a phylogenetic classification at the sectional and species levels has not yet been achieved; therefore, the authors adhere to a more traditional, morphology-based concept in the classification used in this book, with the exception of including the sequestrate genera *Arcangeliella*, *Zelleromyces*, and *Gastrolactarius* that have previously been shown to be synonymous with *Lactarius*.

The authors note that approximately 25% of the species described in this volume are known only from the type locality, highlighting the fragmentary state of knowledge about *Lactarius* (the same could be said of most other genera) in tropical Africa; at the same time, however, the present volume makes an extremely valuable contribution toward reducing the size of this problem. While the high endemism of the African mycota reduces somewhat the utility of this monograph for identifying species found elsewhere, the data and specimens represented therein provide a critical component for understanding the biogeography of *Russulaceae* and tropical ectomycorrhizal fungi in general. The detailed introductory section on taxonomically valuable characters alone is an important enough resource that researchers and students of *Lactarius* should own a copy of this book. This impressive volume excels both in terms of scientific value and aesthetic quality, and I highly recommend it not only to

persons with a specific interest in *Lactarius* or the African mycota, but to any amateur or professional mycologist who wishes to be inspired by an outstanding example of taxonomic mycology.

Heim R. 1938. Les lactario-russulés du domaine oriental de Madagascar. Prodr. Fl. Mycol. Madagascar Dépendances 1: 1–196.

Heim R. 1955a. Les lactaires d'Afrique intertropicale (Congo belge et Afrique noire française). Bull. Jard. Bot. Etat Bx. 25: 1–91.

Heim R. 1955b. *Lactarius*. Flore Iconographique des Champignons du Congo 4: 81–97.

Verbeken A, Buyck B. 2002. Diversity and ecology of tropical ectomycorrhizal fungi in Africa. In: Watling R, Frankland JC, Ainsworth AM, Isaac S, Robinson CH. (eds). Tropical Mycology, Volume 1: Macromycetes: 11–24. Wallingford, CABI Publishing.

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## BOOK ANNOUNCEMENTS

***Corticiceaes. I. Fungi Europaei 12.*** By A. Bernicchia & S.P. Gorjón. 2010. Edizioni Candusso, Via Ottone Primo 90, 17021 Alassio SV, Italy. <maxcandusso@libero.it>. Pp. 1008, plates 427, figs 455. ISBN 978-88-901057-9-1. Price: 77.00 €.

**Rare and interesting species of heterobasidiomycetes from Russia.** Fungi non delineati 53. By V.F. Malysheva, 2010. Edizioni Candusso, Via Ottone Primo 90, 17021 Alassio SV, Italy. <maxcandusso@libero.it>. Pp. 90, plates 52, figs 43. Price: 12.00 €.

**The Lichen Genus *Rinodina* (*Lecanoromycetidae*, *Physciaceae*) in North America, North of Mexico.** By J. Sheard, 2010. NRC Research Press, 1200 Montreal Rd, Bldg M-55, Ottawa, ON K1A 0R6, Canada. <pubs@nrcresearchpress.com>. Pp. 246. ISBN-139780660199412. Price: US\$89.95.

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## **Fungal nomenclature.**

### **Summary of recent decisions by the Nomenclature Committee for Fungi**

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**Abstract** — Recent decisions made by the IAPT permanent Nomenclature Committee for Fungi (NCF) cover 17 proposals to conserve or protect fungal names. Recommendations on 10 sets of proposals to amend the International Code of Botanical Nomenclature (including the governance of fungal nomenclature, name deposition, electronic publication, sanctiotypification, and Art. 59) and decisions on two cases of near homonymy and one of orthography are also reported.

In preparation for IBC2011 (the XVIII International Botanical Congress, Melbourne, Australia, 23–30 July, 2011), the IAPT permanent Nomenclature Committee for Fungi (NCF) reports on votes from two ballots on proposals to conserve or reject fungal names and announces recommendations on proposals to amend the INTERNATIONAL CODE OF BOTANICAL NOMENCLATURE to help guide non-mycologists during the pre-Congress paper ballot and final voting at the July 18–22 Nomenclature Section.

The 14 voting NCF members are Lee Crane (Urbana-Champaign IL), Chairman Vincent Demoulin (Liege), David Hawksworth (Madrid/London), Teresa Iturriaga (Caracas), Paul Kirk (Egham), Pei-Gui Liu (Kunming), Tom May (Melbourne), Jacques Melot (Reykjavík), Secretary Lorelei Norvell (Portland OR), Shaun Pennycook (Auckland), Christian Printzen (Frankfurt), Scott Redhead (Ottawa), Svengunnar Ryman (Uppsala), and Dagmar Triebel (München). As a 9-vote minimum is required for the NCF to recommend or reject a conservation proposal, only those proposals showing a greater than 60% majority can be considered to have passed out of Committee.

Published nomenclatural proposals and NCF reports can be downloaded from [www.ingentaconnect.com/content/iapt/tax] (the TAXON website),

while all previous and current NCF commentaries, important committee correspondence, and interim reports are available via the International Mycological Association website [<http://www.ima-mycology.org/CFF>].

### Proposals to conserve or reject fungal names

\* = proposal decisions detailed in Norvell (2011: TAXON 60(1) in press).

The Committee recommends the following proposals:

- \*PROP. 1810, to conserve the name *Hemipholiota* against *Nemecomyces* (Agaricales, Basidiomycota) [Jacobsson & Holec 2008; TAXON 57: 641-642]  
— 86% support
- \*PROP. 1828, to conserve the name *Aspicilia aquatica* against *Lichen mazarinus* (Ascomycota: Pertusariales: Megasporaceae) [Nordin & Jørgensen 2008; TAXON 57: 989]  
— 86% support
- \*PROP. 1831, to conserve the name *Mixia* against *Phytocerationomyxa* (Basidiomycota) [Sugiyama & Katumoto 2008; TAXON 57: 991-992]  
— 86% support
- PROP. 1852, to conserve the name *Olivea tectonae* (T.S. Ramakr. & K. Ramakr.) R.L. Mulder against *Olivea tectonae* (Racib.) Thirum. (Basidiomycota). [Minnis & al. 2008; TAXON 57: 1355-1356]  
— 93% support
- \*PROP. 1862, to conserve the name *Psoroma versicolor* (*Degeliella versicolor*) against *Psoroma subdescendens* (lichenized Ascomycota, Pannariaceae) [Fryday & Coppins 2009; TAXON 58: 293]  
— 86% support
- PROP. 1863, to conserve the name *Craterellus cinereus* (Pers. : Fr.) Donk with a conserved type against *Craterellus cinereus* Pers. (Basidiomycota) [Olariaga & al. 2009; TAXON 58: 294-295]  
— 93% support
- PROP. 1896, to conserve the name *Lichen lichenoides* (*Leptogium lichenoides*) against *Lichen tremelloides* and *L. tremella* (lichenized Ascomycota) [Jørgensen 2009, TAXON 58: 1002-1003]  
— 71% support
- \*PROP. 1897, Proposal to reject the name *Lecidea epiploica* (lichenized Ascomycota) [Jørgensen & Nordin 2009; TAXON 58: 1003-1004]  
— 93% support
- \*PROP. 1898, to conserve *Stirtonia* A.L. Sm, (lichenized Ascomycota, Arthoniales) against *Stirtonia* R. Gr. bis (Bryophyta, Dicranales) [Frisch & Thor 2009; TAXON 58: 1004]  
— 86% support)



- \*PROP. 1899, to conserve the name *Hebeloma cylindrosporum* against *Hebeloma angustispermum* (*Basidiomycota*) [Vesterholt & al. 2009: TAXON 58: 1005]  
— 93% support
- \*PROP. 1918, to conserve the name *Dermatocarpon (Placopyrenium) bucekii* against *Placidium steineri* (lichenized *Ascomycota*, *Verrucariaceae*) [Senkardesler 2010: TAXON 59: 294]  
— 86% support
- \*PROP. 1919, to conserve *Lactarius* (*Basidiomycota*) with a conserved type [Buyck & al. 2010: TAXON 59: 295–296]  
— 79% support
- \*PROP. 1926, to conserve *Cladia* against *Heterodea* (*Ascomycota*) [Lumbsch & al. 2010: TAXON 59: 643]  
— 86% support
- \*PROP. 1945, to conserve the name *Thelephora comedens* (*Vuilleminia comedens*) with a conserved type (*Basidiomycota*) [Ghobad-Nejhad & Hallenberg 2010: TAXON 59: 1277–1278]  
— 100% support

The Committee does not recommend the following proposals:

- PROP. 1769, to conserve the name *Cortinarius speciosissimus* against *C. rubellus*. [Gasparini & al. 2007: TAXON 56: 596–597]  
— 86% oppose
- \*PROP. 1829–30, to reject the names *Verrucaria thelostoma* (1829) and *Pyrenula umbonata* (1830) (lichenized *Ascomycota*) [Jørgensen 2008: TAXON 57: 990–991]  
— Both opposed: (1829) by 71%; (1830) by 79%

The Committee is still considering the following proposals:

- PROP. 1861, to conserve the name *Aspicilia farinosa* (*Ascomycota*: *Pertusariales*: *Megasporaceae*) with a conserved type [Nordin & Roux 2009: TAXON 58: 292]
- PROP. 1888, to conserve the name *Glomus* (*Fungi*, *Glomeromycota*, *Glomerales*) as being of neuter gender [Kuyper 2009: TAXON 58: 647]  
—NOTE: 93% support the proposal, which is retained for further discussion by request of Chair Demoulin.
- PROP. 1927, to conserve the name *Agaricus rachodes* (*Basidiomycota*) with that spelling [Vellinga & Pennycook 2010: TAXON 59: 644]

## Proposals to amend the International Code of Botanical Nomenclature

The following recommendations cover proposals unrelated to Art. 59:

- PROPS. 16–20, to make clear that the *Code* covers the nomenclature of fungi, and to modify its governance with respect to names of organisms treated as fungi

[Hawksworth & al. 2009: TAXON 58: 658–659]

- 78% support (16—changing the title to the International Code of Botanical and Mycological Nomenclature) and 71% support Props. (17—replacing “plant/s” by “plant/s or fungus/i” throughout) and (18—provide for election of the NCF by an International Mycological Congress). At the moment simple majorities do not support either (19—to permit decisions on fungal proposals to be taken at an IMC) or (20—to make such decisions binding on the subsequent IBC Nomenclature section.)

PROPS. 48–51, to exclude the phylum *Microsporidia* from the *Code* [Redhead & al. 2009: TAXON 58: 669]

- 86% support all three proposals.

PROPS. 117–119, to make the pre-publication deposit of key nomenclatural information in a recognized repository a requirement for valid publication of organisms treated as fungi under the Code [Hawksworth & al. 2010: TAXON 59: 1297]

- 79% support all three proposals.

PROPS. 183–184, to require deposition of information concerning typification of names of fungal taxa, with an associated Recommendation [Gams 2010: TAXON 59: 1626–1627]

- 72% support both proposals

PROPS. 185–190, to amend Art. 15 (185—to clarify what is meant by sanctioning), Art. 36 (185–189—to permit the use of either Latin or English for valid publication), and to amend Art. 45 (190—to make Art. 45 applicable to groups similar to the *Microsporidia* but which are not covered by Props. 48–51) [Demoulin 2010, TAXON 59: 1627–1628]

- All supported: (185) by 86%; (186–189) by 79%; (190) by 71%.

PROPS. 203–213, to permit electronic publications to be effectively published under specified conditions [Special Committee on Electronic Publication 2010: TAXON 59: 1907]

- 79% support

PROP. 223–232, to amend articles regulating the typification of names in sanctioning works [Redhead & al. 2010: TAXON 59: 1910–1913]

- 71% do not support (223—delete Art. 7.8); a 57% simple majority supports (223–232—amend Art. 7.8) .

The following recommendations cover Art. 59 proposals:

PROPS. 172–174, to amend Article 59 concerning teleotypification of fungal names. [Gams & al. 2010: TAXON 59: 1297]

- 71% do not recommend (172) to delete Art. 59.7 and 64% do not support (174) to add Rec. 59A4 to classify a new anamorph under a teleomorph-typified generic name only when no suitable anamorph-typified generic name is available; (173), to alter Art. 59.7 so that teleomorph-typified names

in anamorphic genera need not be changed, is still under consideration with 57% currently opposing.

PROPS. 294–306, to define the new term ‘teleotype’ (294–5), to rename Chapter VI (306), and to modify Art. 59 to limit dual nomenclature and to remove conflicting examples and recommendations (296–305) [Redhead 2010: TAXON 59: 1927–1929]

- A strong majority (64–86%) supports all except 298, 300, and 303; the last three show majority (57%) support.

PROPS. 307–313, to harmonize Art. 59 in order to harmonize it with present practice, by raising the status of anamorph names (307–309), clarify the status of teleomorph- and anamorph-typified genera (310–311), and recommend that teleomorph-typified genera should be reserved to teleomorph-typified species and vice versa for anamorphs (312–313) [Gams & al. 2010: TAXON 1929–1930]

- All proposals are still under consideration. simple majorities support (307–57%) and do not support (308, 310–313—50%); there is no agreement on (309).

### Other recommendations

The following recommendations cover near homonymy according to Art. 53.5 (1–2) and orthography (3).

(1) *Calongea* Healey & al. in *Anales Jard. Bot. Madrid* 66(51): 27. 2009 (*Pezizaceae*) and *Calongia* D. Hawksw. & Etayo in *Lichenologist* 42: 355–359. 2010 (mitosporic fungi).

- 93% considered the names are sufficiently alike to be confusable, and so they should be treated as homonyms, with priority granted to *Calongea* Healey & al.

(2) *Phyllocratera* Sérus. & Aptroot in Aptroot & al., *Biblioth. Lichenol.* 64: 132. 1997 (*Phyllobatheliaceae*) and *Phyllocrater* Wernham in *J. Linn. Soc., Bot.* 42: 90. 1914 (*Dicotyledones, Rubiaceae*).

- 64% considered the names are sufficiently alike to be confusable, and so they should be treated as homonyms, with priority granted to *Phyllocrater* Wernham. (The lower support in case (2) is attributable that two different kingdoms (*Fungi* vs. *Plantae*) are represented.

(3) Regarding the applicability of Art. 60.1 to the elements ‘rhiz,’ ‘rrhiz,’ ‘riz,’ or ‘rriz’ within a name:

- 86% considered that the element should be spelled as written by the original author. Demoulin’s Prop. 185 to amend the Code is an outgrowth of this discussion.



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*Alboleptonia cystidiosa* Largent & Aime, p. 120  
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    J.D. Rogers 1998  
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## FROM THE *EDITOR-IN-CHIEF*

FAREWELL TO HARD COPY — It is with a certain amount of regret I close MYCOTAXON 114 — a volume delayed by a perfect storm of computer and other problems. Ever since I saw my first volume in the 1970's, I have loved the 'book' feel of these brightly colored volumes dedicated solely to fungal nomenclature and taxonomy. While I will not miss compiling author indices or explaining for the umpteenth time why a line drawing must have 900-1200 dpi resolution (or why color — which conveys so much more than halftones ever can — costs so much), I will definitely miss the solid thump of a newly arrived volume on my desktop and sitting down to read all of its pages over again in one continuous flow.

Fortunately, the searchability, versatility, and immediacy of an online journal will more than make up for the sound of that gratifying thump, just as the free and unlimited number of color plates on the 'inside' pages will more than compensate for a book bound in color. For MYCOTAXON, "Print is dead. Long live the pixel!"

OPEN ACCESS — "Knowledge is power. Knowledge shared is power multiplied," wrote the late "mayor of Silicon Valley" (Robert Noyce). For that reason MYCOTAXON feels strongly that all scientific papers should be available to everyone at no charge. Although we cannot cover our costs by making all papers freely available at the outset, through CYBERLIBER and soon INGENTA we do release all papers to the Internet for free access after two years. Nonetheless, we urge authors who can afford our modest and reasonable fee of \$20/page to pay for immediate "OPEN ACCESS."

WEB-LIST INNOVATIONS: FAREWELL TO THE 4-PAGE SUMMARY — As it makes no sense whatsoever to post online a summary of a longer annotated species distribution list ("web-list") also posted online, we no longer require authors to prepare both a summary for inclusion within the journal and a longer annotated list for posting to the MYCOTAXON website. Instead, we now ask that each annotated web-list undergo vigorous and thorough reviews by at least

THREE experts, one of whom is a native English-speaker. After three experts have returned favorable reviews (accompanied by a special 'list' review form) to both authors and *Editor-in-Chief*, the authors may prepare their document using whatever format and size they prefer before submitting it to the *Nomenclature Editor* for accessioning and approval (but not review). Authors then submit their approved, final list (as document or PDF file) + new "list submission form" to the *Editor-in-Chief*. The authors and title of a finally approved list will be cited on a free access summary page within the online volume. The page will list all newly uploaded weblists, each of which will be 'hot-linked' to the MYCOTAXON weblist page. Our weblist upload fee remains \$40. We now also charge \$40 to replace a previously posted species list with an updated and revised version.

NEW INSTRUCTIONS — With the delay of MYCOTAXON 114 and additional time needed to prepare for an online MYCOTAXON 115, I have not yet had time to revise the Instructions to Authors PDF posted on MYCOTAXON website. I have, however, been able to prepare newly revised templates, a sample manuscript, and forms, all of which can be downloaded from the AUTHOR DOWNLOADS PAGE on our website. As noted above, we now require separate weblist review and submission forms. Also, all illustration files should be submitted in JPG (or TIF) format and all should have 300 dpi resolution for a 4.33 page width. Only plates intended to display color should be submitted in color mode.

Warm (if seriously belated) regards,

Lorelei Norvell,  
MYCOTAXON *Editor-in-Chief*  
24 January 2011

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Zhou, Na, see Chen & al.

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ERRATA

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p. 257, line 7	for: <i>Dac. phymatopagum</i>	read: <i>Dac. phymatopaga</i>
line 10	for: <i>Dac. ellipsosporum</i>	read: <i>Dac. ellipsospora</i>
line 11	for: <i>Dac. haptotylum</i>	read: <i>Dac. haptotyla</i>
p. 285, line 12	for: <i>Crepidotus betula</i>	read: <i>Crepidotus betulae</i>
p. 333, line 36	for: <i>Metatrichia vesparia</i>	read: <i>Metatrichia vesparium</i>
p. 336, line 25	for: <i>Metatrichia vesparia</i>	read: <i>Metatrichia vesparium</i>
p. 337, line 34	for: <i>vesparia</i>	read: <i>vesparium</i>
p. 338, line 48	for: <i>Metatrichia vesparia</i>	read: <i>Metatrichia vesparium</i>
p. 340, line 48	for: <i>Metatrichia vesparia</i>	read: <i>Metatrichia vesparium</i>
p. 348, line 6	for: <i>Metatrichia vesparia</i>	read: <i>Metatrichia vesparium</i>
p. 350, line 6	for: <i>Metatrichia vesparia</i>	read: <i>Metatrichia vesparium</i>
p. 352, line 15	for: <i>Metatrichia vesparia</i>	read: <i>Metatrichia vesparium</i>

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p. 455, tab 3	for: <i>Xeroceps peckianus</i>	read: <i>Xanthoporus peckianus</i>
	for: <i>Xeroceps syringae</i>	read: <i>Xanthoporus syringae</i>

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p. 14, line 2	for: nine	read: eight
p. 58, lines 25 & 28	for: Botanik	read: Botanisk
p. 248, line 6	for: <i>B. zelandiaenovae</i>	read: <i>B. zelandiae-novae</i>
p. 393, right_line 4	for: Hyphodiscus-Catinifera	read: Hyphodiscus-Catenulifera
p.468, FIG. 1, line 5	for: culture on wood; show the ...	read: culture on wood show the ...
p.468, bottom line	for: 12 µm long); 1.5 µm wide	read: 12 µm long; 1.5 µm wide
p.469, line 8	for: droplets). Ascospores ...	read: droplets. Ascospores ...
p. 505, 5 <sup>th</sup> from bottom	for: Index of Fungi	read: Index Fungorum
p.505, 2 <sup>nd</sup> from bottom	for: Rec. 88B.3	read: Rec. 8B.3

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